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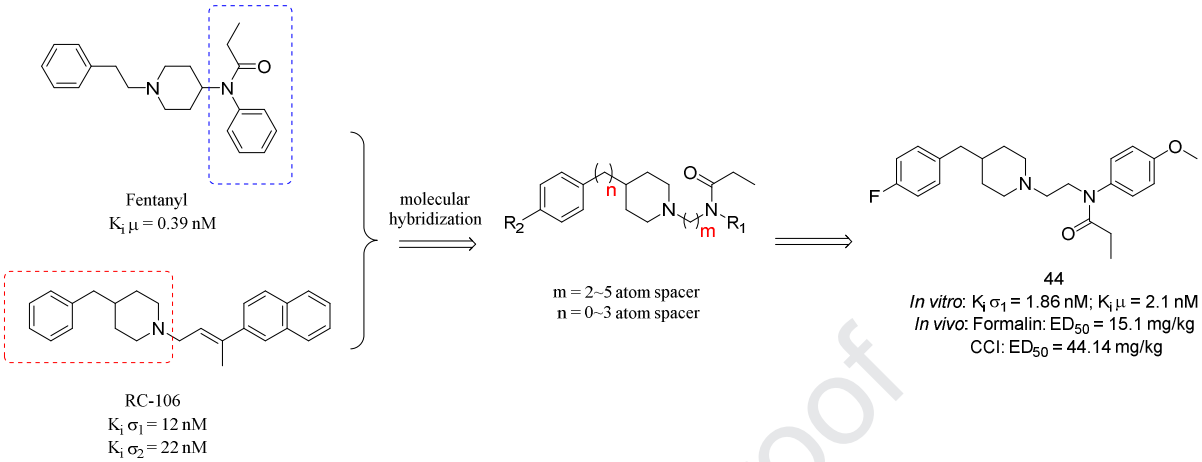
Graphical Abstract



Piperidine propionamide as a scaffold for potent sigma-1 receptor antagonists and mu opioid receptor agonists for treating neuropathic pain

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Keywords

Piperidine propionamide; Sigma-1 receptor antagonists; Mu receptor agonists; Analgesic

Abstract

We designed and synthesized a novel series of piperidine propionamide derivatives as potent sigma-1 (σ_1) receptor antagonists and mu (μ) opioid receptor agonists, and measured their affinity for σ_1 and μ receptors *in vitro* through binding assays. The basic scaffold of the new compounds contained a 4-substituted piperidine ring and N-aryl propionamide. Compound 44, N-(2-(4-(4-fluorobenzyl) piperidin-1-yl) ethyl)-N-(4-methoxy-phenyl) propionamide, showed the highest affinity for σ_1 receptor (K_i σ_1 = 1.86 nM) and μ receptor (K_i μ = 2.1 nM). It exhibited potent analgesic activity in the formalin test (ED_{50} = 15.1 ± 1.67 mg/kg) and had equivalent analgesic effects to S1RA (σ_1 antagonist) in a CCI model. Therefore, Compound 44, which has mixed σ_1/μ receptor profiles, may be a potential candidate for treating neuropathic pain.

1. Introduction

Neuropathic pain is an expensive and debilitating condition that affects 7%–10% of the general population, with the prevalence increasing with age [1]. First-line treatments for neuropathic pain include anticonvulsants, tricyclic antidepressants,

serotonin-norepinephrine reuptake inhibitor antidepressants. In addition, opioids provide second-line treatment. Unfortunately, these drugs are limited in their efficacy and have side effects, in particular with chronic use [2, 3]. Opioids, one category of analgesics, can rapidly reduce symptoms in patients with severe neuropathic pain [4]; however, long-term use can lead to respiratory depression, constipation, tolerance, and physical dependence [5]. Thus, there is a need to explore novel analgesic mechanisms that improve the efficacy of existing therapies while reducing their adverse effects.

In 1976, Martin discovered sigma receptors, which were initially thought to be a new subtype of opioid receptor [6]. Six years later, Su and colleagues demonstrated that sigma receptors differ from opioid receptors [7]. σ_1 receptor, a subtype of sigma receptor, is a chaperone protein that modulates the activity of the NMDA receptor; opioid receptors; and several ion channels, including the K^+ and Ca^{2+} channels [8, 9]. Many studies have validated the involvement of σ_1 receptor in moderating pain. Cendán found that σ_1 receptor knockout mice exhibited reduced pain sensitivity in the formalin test [10]. **Haloperidol (1)**, a nonselective σ_1 receptor antagonist, inhibits capsaicin-induced mechanical hypersensitivity [11, 12]. **S1RA (2)**, a highly active and selective σ_1 receptor antagonist, exhibits analgesic activity in neuropathic pain models [13] and is currently in Phase II clinical trials for treating neuropathic pain. Moreover, ligands of σ_1 receptor antagonists with heterocyclic rings of **pyrimidine (3)**, **pyridazinone (4)**, and **1,2,4-oxadiazole (5)** scaffolds have been designed and tested. Each ligand exhibits dose-dependent analgesic effects in pain models (Fig. 1) [14-16]. Despite these findings, there are currently no selective σ_1 receptor antagonists analgesics on the market.

σ_1 receptor was originally being used for analgesia when Chien and Pasternak identified its role in the endogenous anti-opioid system. σ_1 receptor agonists counteract opioid receptor-mediated analgesic effects, whereas σ_1 receptor antagonists, such as haloperidol, have potentiating effects [17, 18]. A series of experiments were conducted with the highly selective σ_1 antagonist S1RA and σ_1R-KO mice to investigate the potentiation of opioid analgesia through inhibiting σ_1 receptor. The results were consistent with those of nonselective σ_1 antagonists [19-21], which suggests that the analgesic mechanism of opioids is related to σ_1 receptor. Therefore, synergistic administration of σ_1 antagonists with

opioid analgesics may enhance the analgesic potency of opioids. Nevertheless, few analgesics with the dual targets of σ_1 and μ receptors have been reported, and none has entered clinical trials to date.

Carroll and colleagues evaluated the biological activity of phenazocine enantiomers *in vitro* and showed that both **(-)-phenazocine (6a)** and **(+)-phenazocine (6b)** bind to σ_1 and μ receptors (Fig. 2) [22]. In addition, phenazocine enantiomers have demonstrated affinity for other opioid receptors [23, 24]. In regard to analgesic potency, the analgesic effects of (+)-phenazocine (ED_{50} = 6.7 mg/Kg) were comparable to those of morphine (ED_{50} = 2.1 mg/Kg) in the hot plate test, whereas (-)-phenazocine was nearly 20 times more potent than morphine (ED_{50} = 0.11 mg/Kg) [25, 26]. Prezzavento found that phenazocine enantiomers acted as strong analgesics in mechanical pain models, which have mixed σ_1 receptor antagonist and μ receptor agonist profiles [24]. Recently, Mónica García proposed using a merging strategy to design a new series of 1-oxa-4,9-diazaspiro [5.5] undecane derivatives **(7)** (Fig. 2) as potential dual ligands for σ_1 and μ receptors [27], given its balanced dual profile and fit to the pharmacophore model. In the mice paw pressure test, Compound 7 (ED_{50} = 15 mg/kg) showed analgesic activity comparable to that of oxycodone (ED_{50} = 4.3 mg/kg) and induced less constipation than oxycodone at equal doses. Thus, these novel compounds with dual pharmacology of σ_1 and μ receptors, such as phenazocine enantiomers and Compound 7 are potential candidates for novel analgesic treatments.

In this study, novel ligands with dual σ_1 and μ receptor pharmacology were explored. N-phenylpropionamide is an important pharmacophore of **fentanyl (8)** and its derivatives, with the piperidine ring acting as the essential group [28, 29]. Rui *et al*. reported that **RC-106 (9)**, which contains 4-benzylpiperidine, exhibits good affinity for both σ_1 and σ_2 receptors (K_i σ_1 = 12 nM, K_i σ_2 = 22 nM) and thus has promise as a novel anticancer drug targeting σ receptors [30]. Here we developed piperidamide derivatives **(10)** using molecular hybridization to link the N-arylpropionamide of fentanyl with the 4-benzylpiperidine of RC-106 (Fig. 3). As a σ_1 receptor antagonist and μ receptor agonist, compound 44 exhibits basic pharmacophore features, including hydrogen bond acceptors (green), hydrophobic groups (blue), positively ionizable groups (red), and hydrophobic aromatic rings (orange-blue; Fig. 4). Which is consistent with the pharmacophore model

proposed by Mónica García [27].

In this study, we synthesized a series of novel compounds of piperidamide derivatives, which were used in structure–activity relationship (SAR) studies evaluating their pharmacological efficacy and in competitive receptor binding assays to determine their relative affinity for σ_1 and μ opioid receptors *in vitro*. These analyses revealed that Compound 44 exhibited high affinity for σ_1 and μ receptors and displayed dose-dependent anti-nociceptive effects in the formalin test and potent analgesic activity in a CCI model. Therefore, Compound 44 is a hit compound with a mixed σ_1/μ receptor ligand that may be useful for treating neuropathic pain.

2. Chemistry

The reaction pathways for the synthesis of the novel compounds studied are outlined in Schemes 1. We synthesized 2-chloro-N-phenylacetamide derivatives 12a–12k by substituting different aromatic amines 11a–11k with chloroacetyl chloride (Scheme 1). Subsequently, a substitution reaction with 4-benzylpiperidine was used to prepare intermediates 13a–13k. The reduction reaction with intermediates 13a–13k using $LiAlH_4$ as a reducing agent in anhydrous tetrahydrofuran generated intermediates 14a–14k. We prepared Compounds 15–25 (Scheme 1, Table 1) by reacting 14a–14k with propionyl chloride, using triethylamine as a base in dichloromethane (DCM) under an ice bath. We synthesized Compounds 31–36 (Scheme 1, Table 2) by reacting 4-methoxyaniline (11b) with 3-chloropropionyl chloride (4-chlorobutyryl chloride or 5-chlorovalerylchloride) and then coupling it with 4-substituted piperidine following the same reaction pathway. Intermediates 12b–12d reacted with 4-(4-methylbenzyl) piperidine or 4-(4-fluorobenzyl) piperidine to obtain the target Compounds 43–48 (Scheme 1, Tables 3–5). Finally, to reduce the oiliness of the compounds, we converted all target products into oxalate for more convenient pharmacological experiments. The free base was dissolved in ethyl acetate, 1.05 equivalents of oxalic acid dihydrate were added, and the mixture was stirred at room temperature for 4 hours to generate oxalate of target compounds.

3. Results and discussion

3.1 Receptor affinity and SAR analyses

The novel series of 4-benzyl piperidamide derivatives were designed according to the

pharmacophoric characteristics of σ_1 receptor and μ receptor ligands. The parent structure of the compounds (Fig. 3) were devised to ensure that the 4-substituted piperidine ring and propionamide acted as the primary scaffolds and to maintain the effects of different arylamines. We investigated all synthesized compounds with radioligand binding assay to analyze their affinity for σ_1 and μ receptors. Some of the tested compounds had mixed nanomolar affinity for σ_1 and μ receptors *in vitro* (Tables 1–5).

We investigated the influence of phenylamino and other various arylamines on affinity for σ_1 and μ receptors (Table 1, Compounds 15–25). Phenylamino group (Compound 15) showed high μ receptor affinity (K_i μ = 3.7 nM) and moderate σ_1 affinity (K_i σ_1 = 38.6 nM). These results may be attributed to the presence of N-phenylpropionamide, a common group of fentanyl and its derivatives. It is interesting that Compounds 16–18, with methoxyl, methyl, or fluorine substituted at the 4-position of phenylamino group, all bound to σ_1 receptor with high affinity, but had slightly decreased affinity for μ receptor, which suggests that the changes to phenylamino group are negligible (Compound 16: K_i σ_1 = 1.5 nM, K_i μ = 20.4 nM; Compound 17: K_i σ_1 = 1.6 nM, K_i μ = 16.7 nM; Compound 18: K_i σ_1 = 4.1 nM, K_i μ = 19.4 nM). Compound 19, which contained 3,4,5-trifluoro-phenylamino group, had weakened σ_1 receptor binding affinity and did not bind to μ receptor. Compound 20 (K_i σ_1 = 402 nM, K_i μ = 238 nM), which contained a pyridine ring in place of the benzene ring in Compound 15, had attenuated affinity for the two types of receptor. We used 3-fluoropyridin-2-amine and 6-(trifluoromethyl) pyridin-2-amine to synthesize Compounds 21 and 22 to investigate reduced activity of the pyridine ring. Compound 21 (K_i σ_1 = 506 nM, K_i μ = 253 nM) had similar affinity for both σ_1 and μ receptors as Compound 20, whereas Compound 22 had no affinity for either receptor. Compounds 23 and 24, which included additional carbon atoms between the benzene ring and the amino group, each bound moderately to σ_1 receptor (Compound 23: K_i σ_1 = 17.4 nM, Compound 24: K_i σ_1 = 18.7 nM) and had low affinity for μ receptor. In Compound 25, 2-phenylethylamino group replaced the phenylamino group, resulting in lower σ_1 receptor binding affinity (K_i σ_1 = 269 nM) and no affinity for μ receptor, likely because of the length of the benzene ring and the amino group linker.

These findings indicate that the N-phenylpropionamide derivatives have the strongest

affinity for σ_1 and μ receptors, with Compound 16 showing the highest σ_1 receptor binding affinity (K_i σ_1 = 1.5 nM) and moderate μ receptor affinity (K_i μ = 20.4 nM; Table 1). These analyses indicate that the spatial distance between phenylpropanamide and the piperidine ring has mildly weakened the binding affinity for σ_1 receptor, and significantly decrease the affinity for μ receptor (Compound 31: K_i σ_1 = 2.2 nM, K_i μ = 43 nM; Compound 32: K_i σ_1 = 18 nM, K_i μ = 78 nM; Compound 33: K_i σ_1 = 42 nM, K_i μ = 165 nM; Table 2). The effects of linker length between phenyl and the piperidine ring were explored. Compound 34 (K_i σ_1 = 39 nM, K_i μ = 26 nM), which substituted 4-benzylpiperidine with 4-phenylpiperidine, bound with moderate affinity to both receptors. Compound 35 (K_i σ_1 = 372 nM, K_i μ = 82 nM) and Compound 36 (K_i σ_1 = 893 nM, K_i μ = 74 nM), σ_1 activity was significantly reduced and bound with moderate affinity to μ receptor as the carbon chain grew. We found that 4-phenylpiperidine group was the optimal parent structure, and two carbon atoms between the piperidine ring and amide group was the optimal linker length.

Because 4-benzylpiperidine and phenylpropanamide were linked by two carbon atoms and showed strong affinity for both σ_1 and μ receptors. Analogs with different substituents at the para position of the benzene ring were synthesized. The 4-benzylpiperidine of Compound 16 was substituted with 4-(4-methylbenzyl) piperidine or 4-(4-fluorobenzyl) to produce Compounds 43 and 44 (Table 3). Compound 43 (K_i σ_1 = 35 nM, K_i μ = 77 nM) had slightly weakened affinity for σ_1 and μ receptors, whereas Compound 44 (K_i σ_1 = 1.86 nM, K_i μ = 2.1 nM) maintained high affinity for σ_1 receptor and bound strongly to μ receptor, which suggests that the electron-withdrawing fluorine atoms may have yielded improved activity. Similarly, Compounds 45 and 46 were derived from Compound 17 (Table 4) and Compounds 47 and 48 were derived from Compound 18 (Table 5). The *in vitro* affinity of Compound 45 (K_i σ_1 = 29 nM, K_i μ = 82 nM) reduced mildly from that of Compound 17. Moreover, Compound 46 (K_i σ_1 = 2.4 nM, K_i μ = 27.9 nM) varied marginally for both receptors. Compound 47 (K_i σ_1 = 47 nM, K_i μ = 148 nM), which contained 4-(4-methylbenzyl) piperidine, exhibited reduced affinity compared to Compound 18. However, Compound 48 (K_i σ_1 = 1.3 nM, K_i μ = 5.6 nM), which contained 4-(4-fluorobenzyl) piperidine and 4-fluorophenyl propenamide, exhibited high affinity for both receptors.

3.2 Selectivity profile

Because Compounds 44 and 48 possessed high affinity for σ_1 and μ, their selectivity with other receptors related to pain (e.g., σ_2, serotoninergic-1A, serotoninergic-2A, H_3, CB_1, CB_2) was investigated with an *in vitro* binding assay (summarized in the supporting information). Compounds 44 and 48 bound weakly to all other receptors (inhibition at 1 μM < 50%; Table 6).

3.3 Acute toxicity

Compounds 44 and 48 bound to σ_1 and μ receptors with high affinity and demonstrated high selectivity compared to other receptors. An acute toxicity test was performed subcutaneously (s.c.) to assess the safety profiles (median lethal dose, LD_{50}) of these compounds (Table 7). Compounds 44 (LD_{50} = 396.7 mg/kg) and 48 (LD_{50} = 415.8 mg/kg) yielded a higher LD_{50} value than S1RA (LD_{50} = 357.4 mg/kg).

3.4 Formalin test

The antiallodynic activity of Compounds 44 and 48 was investigated with the formalin test. Formalin-induced nociceptive behavior in mice is characterized by a typical acute chemical nociceptive pain response (Phase I) and subsequent persistent pain response (Phase II). The acute pain response results from the stimulation of subcutaneous chemoreceptors and Group C nerve fibers that transmit the pain signal. The secondary pain response is the combined result of central sensitization of the pain stimulus and a peripheral inflammatory response. We assessed the pain response in mice by measuring the time spent licking or biting the injected paw after injection[31, 32].

Mice were pretreated with Compounds 44 and 48 or S1RA at a dose of 50 mg/kg (s.c.) 30 min before the formalin injection. The S1RA (50 mg/kg s.c.) treatment group showed a negligible decline in licking/biting time in Phase I and 273 ± 12.15 s spent licking/biting in Phase II. Compound 44 inhibited the formalin-induced nociceptive behavior more effectively, reducing licking/biting time to 21 ± 3.25 s in Phase I and 63 ± 4.53 s in Phase II. Compound 48 reduced licking/biting time to 36 ± 3.54 s and 110 ± 9.47 s in Phase I and Phase II, respectively (Fig. 5).

The analgesic effects of Compound 44 were further investigated at different doses (12.5–100 mg/kg). In Phase II, licking/biting time decreased significantly as the dose

increased (201 ± 12.86 s, 108 ± 7.61 s, 63 ± 4.53 s, and 41 ± 2.31 s at 12.5, 25, 50, and 100 mg/kg s.c., respectively), demonstrating dose-dependent analgesic effects (Fig. 6). The ED_{50} value was 15.1 ± 1.67 mg/kg for Phase II in the formalin test. The therapeutic index (TI = LD50/ED50) of Compound 44 in the formalin test was 26.27, which indicates improved safety compared to S1RA (TI = 11.98, in previous studies).

3.5 CCI model

The chronic constriction injury (CCI) model is the most widely used tool for studying neuropathic pain [33], as it allows investigators to study the main characteristics of neuropathic pain, including spontaneous pain, hyperalgesia, and allodynia [34]. The CCI model involves mechanical hyperalgesia and is relatively stable. The mechanical withdrawal threshold (MWT) was measured as an indicator of mechanical allodynia. Compound 44 (25 mg/kg) was evaluated in a rat CCI model of neuropathic pain and yielded an analgesic effect equivalent to that of S1RA (50 mg/kg; Fig. 7). At day 15, Compound 44 showed dose-dependent inhibition of mechanical allodynia with a single-dose treatment, with an ED_{50} value of 44.14 ± 3.25 mg/kg.

4. Conclusion

This report details the synthesis of and structure–activity relationships among a novel series of piperidine propionamide derivatives. Several factors affected the binding affinity for σ_1 and μ receptors in this family of compounds. The results showed that phenylamino group or para-substituted phenylamino group were the optimal scaffold and two carbon units was the most suitable linker length between phenylpropanamide and the piperidine ring. In addition, the 4-(4-fluorobenzyl) piperidine showed the most potent σ_1/μ receptor affinity. Of all compounds studied, Compound 44 exhibited the highest affinity for σ_1 and μ receptors and an excellent selectivity profile. Compound 44 showed dose-dependent analgesic effects in Phase II of the formalin test and exhibited anti-neuropathic pain effects consistent with S1RA in the CCI model. These findings suggest that Compound 44 may be a potential candidate drug for treating neuropathic pain.

5. Experimental

5.1 Chemistry experimental

All commercially available chemicals and reagents were used without further

purification. Reagents were all of analytical grade (>99%) or of chemical purity (>95%). Melting points were determined on a Mel-TEMP II meltingpoint apparatus and are uncorrected. ^{1}H and ^{13}C NMR spectra were recorded on a Bruker Avance III 400 spectrometer at 400 MHz (^{1}H) and 101 MHz (^{13}C) or Bruker Avance III 600 spectrometer at 600 MHz (^{1}H) and 151 MHz (^{13}C) using $CDCl_3$ as solvent. Chemical shifts were given in δ values (ppm), using tetramethylsilane (TMS) as the internal standard; coupling constants (J) were given in Hz. Signal multiplicities were characterized as s (singlet), d (doublet), t (triplet), q (quartet), m (multiplet), br (broad signal). Analytical thin layer chromatography (TLC) was performed on silica gel GF254. Column chromatographic purification was carried out using silica gel. Compound purity is determined by high performance liquid chromatography (HPLC), and all final test compounds were >95% purity.

HPLC methods used the following: Agilent 1260 Infinity II spectrometer; column, Inert Sustain C18 5.0 mm × 150 mm × 4.6 mm i.d. (SHIMADZU, Japan); mobile phase, 30 mmol NH_4COOH (ROE SCIENTIFIC INC, USA) aq/acetonitrile (Merck Company, Germany) 45/55; flow rate, 1.0 mL/min; column temperature, 40 °C. Compounds 12a-12d [35], 12f [36], 12i [37], 12j-12k [38], 26a [39], 26b [40] were prepared according to the reported procedures.

5.1.1 General method for preparation of compounds 12e, 12g, and 12h [35].

10.0 mmol of 3, 4, 5-trifluoroaniline, (or 3-fluoropyridin-2-amine or 6-(trifluoromethyl) pyridin-2- amine), and potassium carbonate (20mmol) was dissolved in 50 mL acetone, and stirred in an ice bath to 0 °C. Chloroacetyl chloride (12.0mmol) was slowly added dropwise and stirred at room temperature for 4 hours. The reaction mixture was quenched with water and extracted with ethyl acetate. The organic layer was washed with water and brine successively, dried over anhydrous sodium sulfate, and concentrated in vacuum. The resulting residue was purified by silica gel chromatography (petroleum ether/EtOAc = 10/1) to give the desired product 12e, 12g and 12h.

5.1.1.1 2-chloro-N-(3, 4, 5-trifluorophenyl) acetamide (12e).

Yield: 86%.Beige solid, m. p. 115-119 °C.^{1}H NMR (400 MHz, $CDCl_3$) δ 8.33 (s, 1H), 7.43 – 7.21 (m, 2H), 4.19 (s, 2H).^{13}C NMR (101 MHz, $CDCl_3$) δ 164.10, 152.43, 152.38, 152.33,

152.28, 149.96, 149.91, 149.86, 149.81, 138.25, 135.77, 132.29, 132.24, 104.70, 104.45, 42.66. MS (ESI) m/z 224.2 (calcd 224.0 for $C_8H_6ClF_3NO^+$ $[M + H]^+$).

5.1.1.2 2-chloro-N-(3-fluoropyridin-2-yl) acetamide (12g).

Yield: 79%. White solid, m. p. 129-132 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.27 (s, 1H), 7.80 (d, *J* = 4.7 Hz, 1H), 7.37 – 7.31 (m, 1H), 6.72 – 6.67 (m, 1H), 3.68 (s, 2H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 168.53, 148.31, 145.81, 139.17, 139.12, 122.98, 122.83, 113.35, 113.32, 29.69. MS (ESI) m/z 189.2 (calcd 189.0 for $C_7H_7ClFN_2O^+$ $[M + H]^+$).

5.1.1.3 2-chloro-N-(6-(trifluoromethyl) pyridin-2-yl) acetamide (12h)

Yield: 81%. Pale yellow solid, m. p. 123-126 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.95 (s, 1H), 8.42 (d, *J* = 8.4 Hz, 1H), 7.92 (t, *J* = 8.0 Hz, 1H), 7.48 (d, *J* = 7.6 Hz, 1H), 4.24 (s, 2H).^{13}C NMR (101 MHz, $CDCl_3$) δ 164.90, 150.55, 146.72, 146.37, 139.96, 122.35, 119.62, 116.89, 116.86, 116.77, 42.65.MS (ESI) m/z 239.2 (calcd 239.0 for $C_8H_7ClF_3N_2O^+$ $[M + H]^+$).

5.1.2 General method for preparation of compounds 13a-13k.

A mixture of intermediate 12a (10mmol) (12b-12k) and 4-benzyl piperidine (11.0mmol) in 100 mL acetone was stirred at room temperature for 8 hours in the presence of potassium carbonate (30mmol). After the reaction was completed, it was quenched with water, and extracted with ethyl acetate; the mixture was washed three times with saturated brine and dried over anhydrous sodium sulfate. Distilling solvent under reduced pressure, the crude product was purified by silica gel chromatography (petroleum ether/EtOAc = 1/1) to yield corresponding intermediates 13a-13k.

5.1.2.1 2-(4-benzylpiperidin-1-yl)-N-phenylacetamide (13a).

Yield: 91%. White solid, m. p. 133-135 °C. ^{1}H NMR (600 MHz, $CDCl_3$) δ 9.20 (s, 1H), 7.57 (d, *J* = 7.6 Hz, 2H), 7.34 – 7.31 (m, 2H), 7.28 (t, *J* = 7.5 Hz, 2H), 7.19 (t, *J* = 7.4 Hz, 1H), 7.14 (d, *J* = 7.1 Hz, 2H), 7.10 (t, *J* = 7.4 Hz, 1H), 3.07 (s, 2H), 2.87 (d, *J* = 11.7 Hz, 2H), 2.57 (d, *J* = 7.2 Hz, 2H), 2.18 (td, *J* = 11.8, 2.2 Hz, 2H), 1.69 (d, *J* = 12.7 Hz, 2H), 1.61 – 1.52 (m, 1H), 1.37 – 1.29 (m, 2H). ^{13}C NMR (151 MHz, $CDCl_3$) δ 168.77, 140.23, 137.64, 129.00, 128.93, 128.19, 125.88, 124.01, 119.34, 62.31, 54.23, 43.00, 37.20, 32.47.MS (ESI) m/z 309.2 (calcd 309.2 for $C_{20}H_{25}N_2O^+$ $[M + H]^+$).

5.1.2.2 2-(4-benzylpiperidin-1-yl)-N-(4-methoxyphenyl) acetamide (13b)

Yield: 84%. Brown solid, m. p. 141-143 °C. ^{1}H NMR (600 MHz, $CDCl_3$) δ 9.07 (s, 1H), 7.50 – 7.45 (m, 2H), 7.28 (t, *J* = 7.5 Hz, 2H), 7.20 (t, *J* = 7.4 Hz, 1H), 7.15 (d, *J* = 7.0 Hz, 2H), 6.89 – 6.85 (m, 2H), 3.79 (s, 3H), 3.06 (s, 2H), 2.87 (d, *J* = 11.7 Hz, 2H), 2.57 (d, *J* = 7.2 Hz, 2H), 2.18 (td, *J* = 11.8, 2.2 Hz, 2H), 1.69 (d, *J* = 13.3 Hz, 2H), 1.61 – 1.52 (m, 1H), 1.33 (qd, *J* = 12.3, 3.8 Hz, 2H). ^{13}C NMR (151 MHz, $CDCl_3$) δ 168.50, 156.19, 140.27, 130.91, 129.02, 128.20, 125.90, 121.06, 114.11, 62.24, 55.45, 54.27, 43.02, 37.22, 32.48. MS (ESI) m/z 339.2 (calcd 339.2 for $C_{21}H_{27}N_2O_2^+$ [M + H] $^+$).

5.1.2.3 2-(4-benzylpiperidin-1-yl)-N-(p-tolyl) acetamide (13c)

Yield: 89%. Yellow solid, m. p. 130-131 °C. ^{1}H NMR (600 MHz, $CDCl_3$) δ 9.12 (s, 1H), 7.45 (d, *J* = 8.4 Hz, 2H), 7.28 (t, *J* = 7.5 Hz, 2H), 7.19 (t, *J* = 7.4 Hz, 1H), 7.16 – 7.10 (m, 4H), 3.06 (s, 2H), 2.87 (d, *J* = 11.7 Hz, 2H), 2.56 (d, *J* = 7.2 Hz, 2H), 2.31 (s, 3H), 2.18 (td, *J* = 11.8, 2.2 Hz, 2H), 1.69 (d, *J* = 13.3 Hz, 2H), 1.60 – 1.51 (m, 1H), 1.33 (qd, *J* = 12.3, 3.8 Hz, 2H). ^{13}C NMR (151 MHz, $CDCl_3$) δ 168.61, 140.27, 135.14, 133.58, 129.42, 129.02, 128.20, 125.89, 119.39, 62.31, 54.24, 43.01, 37.22, 32.48, 20.82. MS (ESI) m/z 323.2 (calcd 323.2 for $C_{21}H_{27}N_2O^+$ [M + H] $^+$).

5.1.2.4 2-(4-benzylpiperidin-1-yl)-N-(4-fluorophenyl) acetamide (13d)

Yield: 82%. Brown solid, m. p. 122-124 °C. ^{1}H NMR (600 MHz, $CDCl_3$) δ 9.19 (s, 1H), 7.56 – 7.50 (m, 2H), 7.28 (t, *J* = 7.5 Hz, 2H), 7.20 (t, *J* = 7.4 Hz, 1H), 7.14 (d, *J* = 7.1 Hz, 2H), 7.04 – 6.99 (m, 2H), 3.07 (s, 2H), 2.87 (d, *J* = 11.7 Hz, 2H), 2.57 (d, *J* = 7.2 Hz, 2H), 2.19 (td, *J* = 11.8, 2.2 Hz, 2H), 1.70 (d, *J* = 13.3 Hz, 2H), 1.61 – 1.53 (m, 1H), 1.33 (qd, *J* = 12.3, 3.8 Hz, 2H). ^{13}C NMR (151 MHz, $CDCl_3$) δ 168.72, 159.96, 158.35, 140.20, 133.75, 133.73, 129.01, 128.20, 125.91, 121.04, 120.99, 115.61, 115.46, 62.18, 54.26, 42.99, 37.17, 32.46. MS (ESI) m/z 327.2 (calcd 327.2 for $C_{20}H_{24}FN_2O^+$ [M + H] $^+$).

5.1.2.5 2-(4-benzylpiperidin-1-yl)-N-(3, 4, 5-trifluorophenyl) acetamide (13e)

Yield: 95%. White solid, m. p. 135-137 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 9.29 (s, 1H), 7.32 – 7.27 (m, 4H), 7.20 (dd, *J* = 8.3, 6.3 Hz, 1H), 7.15 (d, *J* = 7.0 Hz, 2H), 3.07 (s, 2H), 2.85 (d, *J* = 11.7 Hz, 2H), 2.58 (d, *J* = 7.1 Hz, 2H), 2.21 (td, *J* = 11.8, 2.2 Hz, 2H), 1.71 (d, *J* = 13.1 Hz, 2H), 1.64 – 1.52 (m, 1H), 1.38 – 1.23 (m, 2H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 169.11, 152.46, 152.35, 150.00, 149.89, 140.13, 133.33, 129.04, 128.25, 125.98, 103.75, 103.49, 99.95, 62.08, 54.30, 42.96, 37.14, 32.43.MS (ESI) m/z 363.3 (calcd 363.2 for

$C_{20}H_{22}F_3N_2O^+$ [M + H] $^+$).

5.1.2.6 2-(4-benzylpiperidin-1-yl)-N-(pyridin-2-yl) acetamide (13f)

Yield: 89%. Brown solid, m. p. 121-123 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 9.67 (s, 1H), 8.33 – 8.29 (m, 1H), 8.26 (d, *J* = 8.4 Hz, 1H), 7.67 (dd, *J* = 11.2, 4.5 Hz, 1H), 7.27 (t, *J* = 7.4 Hz, 2H), 7.20 – 7.11 (m, 3H), 7.04 – 6.99 (m, 1H), 3.09 (s, 2H), 2.86 (d, *J* = 11.5 Hz, 2H), 2.54 (d, *J* = 7.0 Hz, 2H), 2.16 (t, *J* = 11.4 Hz, 2H), 1.65 (d, *J* = 12.6 Hz, 2H), 1.60 – 1.47 (m, 1H), 1.45 – 1.38 (m, 2H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 169.49, 150.92, 147.78, 140.22, 138.07, 128.88, 128.04, 125.71, 119.53, 113.61, 62.33, 54.16, 42.84, 37.10, 32.10. MS (ESI) m/z 310.2 (calcd 310.2 for $C_{19}H_{24}N_3O^+$ [M + H] $^+$).

5.1.2.7 2-(4-benzylpiperidin-1-yl)-N-(3-fluoropyridin-2-yl) acetamide (13g)

Yield: 83%. Purple solid, m. p. 129-131 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 9.77 (s, 1H), 8.28 (d, *J* = 4.6 Hz, 1H), 7.49 – 7.44 (m, 1H), 7.31 – 7.26 (m, 2H), 7.20 (dd, *J* = 8.4, 6.3 Hz, 1H), 7.17 – 7.09 (m, 3H), 3.16 (s, 2H), 2.93 (d, *J* = 11.7 Hz, 2H), 2.56 (d, *J* = 7.1 Hz, 2H), 2.23 (td, *J* = 11.7, 2.2 Hz, 2H), 1.70 (d, *J* = 13.4 Hz, 2H), 1.63 – 1.48 (m, 1H), 1.40 – 1.31 (m, 2H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 168.68, 151.57, 148.99, 143.92, 143.87, 140.29, 140.12, 140.01, 129.01, 128.18, 125.87, 123.79, 123.62, 121.02, 120.99, 62.24, 54.25, 42.98, 37.17, 32.42. MS (ESI) m/z 328.2 (calcd 328.2 for $C_{19}H_{23}FN_3O^+$ [M + H] $^+$).

5.1.2.8 2-(4-benzylpiperidin-1-yl)-N-(6-(trifluoromethyl) pyridin-2-yl) acetamide (13h)

Yield: 75%. White solid, m. p. 109-111 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 9.73 (s, 1H), 8.46 (d, *J* = 8.4 Hz, 1H), 7.85 (t, *J* = 8.0 Hz, 1H), 7.41 (d, *J* = 7.5 Hz, 1H), 7.28 (dd, *J* = 13.8, 6.3 Hz, 2H), 7.21 (d, *J* = 7.3 Hz, 1H), 7.16 (d, *J* = 7.1 Hz, 2H), 3.12 (s, 2H), 2.87 (d, *J* = 11.6 Hz, 2H), 2.59 (d, *J* = 7.0 Hz, 2H), 2.19 (td, *J* = 11.7, 1.8 Hz, 2H), 1.68 (d, *J* = 13.4 Hz, 2H), 1.62 – 1.51 (m, 1H), 1.48 – 1.40 (m, 2H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 170.20, 151.33, 146.57, 146.22, 140.42, 139.51, 129.07, 128.21, 125.89, 122.56, 119.84, 116.74, 115.97, 115.94, 62.55, 54.40, 42.95, 37.32, 32.12. MS (ESI) m/z 378.3 (calcd 378.2 for $C_{20}H_{23}F_3N_3O^+$ [M + H] $^+$).

5.1.2.9 2-(4-benzylpiperidin-1-yl)-N-(1-phenylethyl) acetamide (13i)

Yield: 67%. Yellow solid, m. p. 122-124 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.52 (d, *J* = 8.4 Hz, 1H), 7.36 – 7.22 (m, 7H), 7.18 (t, *J* = 7.3 Hz, 1H), 7.14 – 7.10 (m, 2H), 5.19 – 5.10 (m, 1H), 3.00 – 2.89 (m, 2H), 2.84 – 2.77 (m, 1H), 2.75 – 2.69 (m, 1H), 2.52 (d, *J* = 7.1 Hz,

2H), 2.06 (qd, *J* = 11.7, 2.3 Hz, 2H), 1.66 – 1.57 (m, 2H), 1.56 – 1.50 (m, 1H), 1.49 (d, *J* = 6.9 Hz, 3H), 1.32 – 1.16 (m, 2H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 169.60, 143.30, 140.24, 128.91, 128.51, 128.08, 127.08, 125.82, 125.76, 61.79, 54.13, 47.79, 42.89, 37.14, 32.23, 32.17, 22.02.MS (ESI) m/z 337.2 (calcd 337.2 for $C_{22}H_{29}N_2O^+$ [M + H] $^+$).

5.1.2.10 N-benzyl-2-(4-benzylpiperidin-1-yl) acetamide (13j)

Yield: 88%. Brown oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.57 (s, 1H), 7.33 – 7.28 (m, 2H), 7.26 – 7.21 (m, 5H), 7.15 (t, *J* = 7.3 Hz, 1H), 7.08 (d, *J* = 7.0 Hz, 2H), 4.45 (d, *J* = 6.1 Hz, 2H), 2.98 (s, 2H), 2.76 (d, *J* = 11.6 Hz, 2H), 2.47 (d, *J* = 7.1 Hz, 2H), 2.05 (td, *J* = 11.7, 2.0 Hz, 2H), 1.58 (d, *J* = 13.0 Hz, 2H), 1.53 – 1.44 (m, 1H), 1.20 (qd, *J* = 12.4, 3.7 Hz, 2H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 170.29, 140.02, 138.32, 128.72, 128.33, 127.88, 127.13, 126.98, 125.55, 61.66, 54.02, 42.68, 42.48, 36.89, 31.94. MS (ESI) m/z 323.2 (calcd 323.2 for $C_{21}H_{27}N_2O^+$ [M + H] $^+$).

5.1.2.11 2-(4-benzylpiperidin-1-yl)-N-phenethylacetamide (13k)

Yield: 82%. Brown oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.25 – 7.21 (m, 6H), 7.17 – 7.12 (m, 3H), 7.07 (t, *J* = 7.4 Hz, 2H), 3.50 (q, *J* = 6.6 Hz, 2H), 2.83 (s, 2H), 2.76 (t, *J* = 6.8 Hz, 2H), 2.57 (d, *J* = 11.4 Hz, 2H), 2.44 (d, *J* = 6.9 Hz, 2H), 1.91 (t, *J* = 10.9 Hz, 2H), 1.47 (d, *J* = 13.2 Hz, 2H), 1.43 – 1.34 (m, 1H), 1.04 (qd, *J* = 12.5, 3.6 Hz, 2H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 169.73, 139.64, 138.24, 138.08, 128.40, 128.07, 127.91, 127.52, 126.76, 126.57, 125.75, 125.21, 61.19, 53.60, 53.48, 42.38, 38.99, 36.42, 34.91, 31.59, 31.54. MS (ESI) m/z 337.2 (calcd 337.2 for $C_{22}H_{29}N_2O^+$ [M + H] $^+$).

5.1.3 General method for preparation of compounds 14a-14k.

The intermediate 13a (10mmol) (13b-13k) was dissolved in 100 mL of anhydrous THF and stirred for 15 min in an ice bath. Then, $LiAlH_4$ (20mmol) was added slowly, the mixture was t refluxed at 65 °C for 4 h. The reaction mixture was quenched with ethanol under 0 °C conditions, and extracted with ethyl acetate. The organic layer was washed with 5% NaOH solution, water and brine successively, and then dried over anhydrous Na_2SO_4 and concentrated in vacuum. The resulting residue was purified by silica gel chromatography (petroleum ether/EtOAc = 1/2) to give the desired product 14a-14k.

5.1.3.1 N-(2-(4-benzylpiperidin-1-yl) ethyl) aniline (14a)

Yield: 80%. Brown oil. ^{1}H NMR (600 MHz, $CDCl_3$) δ 7.24 (t, *J* = 7.5 Hz, 2H), 7.17 – 7.13

(m, 3H), 7.11 (d, *J* = 7.0 Hz, 2H), 6.67 (t, *J* = 7.3 Hz, 1H), 6.59 (dd, *J* = 8.5, 0.9 Hz, 2H), 4.27 (s, 1H), 3.09 (t, *J* = 5.7 Hz, 2H), 2.83 (d, *J* = 11.6 Hz, 2H), 2.51 (dd, *J* = 12.7, 6.7 Hz, 4H), 1.87 (td, *J* = 11.8, 2.1 Hz, 2H), 1.59 (d, *J* = 13.2 Hz, 2H), 1.54 – 1.46 (m, 1H), 1.30 – 1.22 (m, 2H). ^{13}C NMR (151 MHz, $CDCl_3$) δ 148.47, 140.49, 129.01, 128.94, 128.00, 125.63, 116.98, 112.73, 56.88, 53.53, 43.05, 40.38, 37.83, 32.07. MS (ESI) m/z 295.2 (calcd 295.2 for $C_{20}H_{27}N_2^+$ $[M + H]^+$).

5.1.3.2 N-(2-(4-benzylpiperidin-1-yl) ethyl)-4-methoxyaniline (14b)

Yield: 71%. Colorless oil. ^{1}H NMR (600 MHz, $CDCl_3$) δ 7.26 – 7.22 (m, 2H), 7.17 – 7.13 (m, 1H), 7.10 (d, *J* = 6.7 Hz, 2H), 6.76 (d, *J* = 8.7 Hz, 2H), 6.71 (d, *J* = 9.2 Hz, 1H), 6.60 – 6.56 (m, 1H), 3.70 (s, 3H), 3.67 (s, 1H), 3.50 – 3.45 (m, 1H), 3.07 (t, *J* = 6.0 Hz, 1H), 2.85 (d, *J* = 11.5 Hz, 2H), 2.54 (tt, *J* = 18.0, 7.5 Hz, 4H), 1.94 – 1.86 (m, 2H), 1.60 (d, *J* = 11.5 Hz, 2H), 1.55 – 1.45 (m, 1H), 1.28 (qd, *J* = 12.4, 3.6 Hz, 2H). ^{13}C NMR (151 MHz, $CDCl_3$) δ 151.83, 142.80, 140.46, 128.92, 127.98, 125.60, 114.68, 113.99, 57.00, 55.59, 53.56, 43.01, 41.40, 37.79, 32.02. MS (ESI) m/z 325.2 (calcd 325.2 for $C_{21}H_{29}N_2O^+$ $[M + H]^+$).

5.1.3.3 N-(2-(4-benzylpiperidin-1-yl) ethyl)-4-methylaniline (14c)

Yield: 68%. Colorless oil. ^{1}H NMR (600 MHz, $CDCl_3$) δ 7.24 (td, *J* = 7.6, 3.5 Hz, 2H), 7.15 (dd, *J* = 12.0, 4.6 Hz, 1H), 7.11 (t, *J* = 6.4 Hz, 2H), 6.97 (d, *J* = 8.2 Hz, 2H), 6.57 – 6.51 (m, 2H), 4.09 (s, 1H), 3.09 (t, *J* = 6.1 Hz, 2H), 2.84 (d, *J* = 11.6 Hz, 2H), 2.56 – 2.49 (m, 4H), 2.22 (s, 3H), 1.88 (td, *J* = 11.8, 2.0 Hz, 2H), 1.60 (d, *J* = 12.9 Hz, 2H), 1.53 – 1.48 (m, 1H), 1.31 – 1.24 (m, 2H). ^{13}C NMR (151 MHz, $CDCl_3$) δ 146.31, 140.53, 129.54, 128.97, 128.03, 126.19, 125.65, 112.99, 56.99, 53.58, 43.08, 40.87, 37.86, 32.09, 20.28. MS (ESI) m/z 309.2 (calcd 309.2 for $C_{21}H_{29}N_2^+$ $[M + H]^+$).

5.1.3.4 N-(2-(4-benzylpiperidin-1-yl) ethyl)-4-fluoroaniline (14d)

Yield: 65%. Brown oil. ^{1}H NMR (600 MHz, $CDCl_3$) δ 7.25 (t, *J* = 7.5 Hz, 2H), 7.15 (td, *J* = 7.1, 4.8 Hz, 2H), 7.11 (t, *J* = 6.5 Hz, 2H), 6.88 – 6.84 (m, 1H), 6.60 (d, *J* = 7.7 Hz, 1H), 6.53 – 6.48 (m, 1H), 4.17 (s, 1H), 3.10 (t, *J* = 6.0 Hz, 1H), 3.04 (t, *J* = 6.0 Hz, 1H), 2.87 – 2.81 (m, 2H), 2.54 – 2.49 (m, 4H), 1.91 – 1.85 (m, 2H), 1.60 (d, *J* = 12.3 Hz, 2H), 1.52 – 1.48 (m, 1H), 1.27 (td, *J* = 14.9, 4.1 Hz, 2H). ^{13}C NMR (151 MHz, $CDCl_3$) δ 148.51, 144.92, 140.53, 140.49, 128.97, 128.03, 125.66, 125.65, 116.99, 115.47, 115.32, 113.50, 113.45, 112.75, 56.90, 56.86, 53.56, 43.07, 41.08, 40.40, 37.85, 37.84, 32.10. MS (ESI) m/z

313.2 (calcd 313.2 for $C_{20}H_{26}FN_2^+$ [M + H] $^+$).

5.1.3.5 N-(2-(4-benzylpiperidin-1-yl) ethyl)-3, 4, 5-trifluoroaniline (14e)

Yield: 72%. Brown oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.27 (t, *J* = 7.4 Hz, 2H), 7.21 – 7.11 (m, 3H), 6.18 – 6.09 (m, 2H), 4.46 (s, 1H), 3.00 (dd, *J* = 11.3, 5.4 Hz, 2H), 2.83 (d, *J* = 11.5 Hz, 2H), 2.53 (dd, *J* = 6.4, 3.7 Hz, 4H), 1.90 (td, *J* = 11.8, 2.1 Hz, 2H), 1.63 (d, *J* = 13.2 Hz, 2H), 1.53 – 1.47 (m, 1H), 1.27 (qd, *J* = 12.5, 3.7 Hz, 2H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 153.17, 153.11, 153.07, 153.00, 150.74, 150.67, 150.63, 150.57, 144.63, 144.51, 144.49, 144.40, 140.55, 133.32, 133.17, 130.95, 130.80, 129.07, 128.13, 125.78, 96.16, 95.92, 56.37, 53.54, 43.12, 40.41, 37.90, 32.15. MS (ESI) m/z 349.2 (calcd 349.2 for $C_{20}H_{24}F_3N_2^+$ [M + H] $^+$).

5.1.3.6 N-(2-(4-benzylpiperidin-1-yl) ethyl) pyridin-2-amine (14f)

Yield: 80%. Brown oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.07 (dd, *J* = 5.0, 1.1 Hz, 1H), 7.41 – 7.35 (m, 1H), 7.27 (dd, *J* = 10.4, 4.3 Hz, 2H), 7.20 – 7.11 (m, 3H), 6.55 – 6.50 (m, 1H), 6.38 (d, *J* = 8.4 Hz, 1H), 5.14 (s, 1H), 3.31 (dd, *J* = 11.5, 5.6 Hz, 2H), 2.88 (d, *J* = 11.6 Hz, 2H), 2.57 – 2.51 (m, 4H), 1.92 (td, *J* = 11.7, 2.1 Hz, 2H), 1.65 – 1.58 (m, 2H), 1.57 – 1.46 (m, 1H), 1.34 – 1.25 (m, 2H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 158.78, 148.04, 140.61, 137.19, 129.03, 128.08, 125.70, 112.47, 106.97, 56.94, 53.61, 43.13, 38.72, 37.90, 32.10. MS (ESI) m/z 296.2 (calcd 296.2 for $C_{19}H_{26}N_3^+$ [M + H] $^+$).

5.1.3.7 N-(2-(4-benzylpiperidin-1-yl) ethyl)-3-fluoropyridin-2-amine (14g)

Yield: 69%. Colorless oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.87 (d, *J* = 5.0 Hz, 1H), 7.30 – 7.25 (m, 2H), 7.19 (dd, *J* = 7.8, 1.6 Hz, 1H), 7.17 – 7.12 (m, 2H), 7.11 – 7.07 (m, 1H), 6.50 – 6.46 (m, 1H), 5.27 (s, 1H), 3.51 (dd, *J* = 11.4, 5.8 Hz, 2H), 2.91 (d, *J* = 11.6 Hz, 2H), 2.59 (t, *J* = 6.1 Hz, 2H), 2.54 (d, *J* = 7.1 Hz, 2H), 1.94 (td, *J* = 11.7, 2.1 Hz, 2H), 1.63 (d, *J* = 13.5 Hz, 2H), 1.59 – 1.48 (m, 1H), 1.36 – 1.26 (m, 2H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 148.89, 148.78, 148.51, 146.01, 142.70, 142.64, 140.71, 129.07, 128.13, 125.75, 119.82, 119.67, 111.43, 111.41, 57.00, 53.64, 43.17, 37.99, 37.70, 32.19. MS (ESI) m/z 314.2 (calcd 314.2 for $C_{19}H_{25}FN_3^+$ [M + H] $^+$).

5.1.3.8 N-(2-(4-benzylpiperidin-1-yl) ethyl)-6-(trifluoromethyl) pyridin-2-amine (14h)

Yield: 58%. Colorless oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.49 (t, *J* = 7.9 Hz, 1H), 7.27 (dd, *J* = 12.5, 5.0 Hz, 2H), 7.21 – 7.12 (m, 3H), 6.89 (d, *J* = 7.3 Hz, 1H), 6.52 (d, *J* = 8.5 Hz,

1H), 5.37 (s, 1H), 3.35 (dd, *J* = 11.6, 5.4 Hz, 2H), 2.88 (d, *J* = 11.6 Hz, 2H), 2.54 (dd, *J* = 9.8, 4.5 Hz, 4H), 1.92 (td, *J* = 11.7, 2.1 Hz, 2H), 1.63 (d, *J* = 13.1 Hz, 2H), 1.58 – 1.51 (m, 1H), 1.36 – 1.25 (m, 2H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 158.59, 146.69, 146.36, 140.64, 137.83, 129.09, 128.14, 125.77, 123.05, 120.33, 110.02, 108.55, 108.51, 56.82, 53.67, 43.17, 38.58, 37.95, 32.12. MS (ESI) m/z 364.2 (calcd 364.2 for $C_{20}H_{25}F_3N_3^+$ [M + H] $^+$).

5.1.3.9 2-(4-benzylpiperidin-1-yl)-N-(1-phenylethyl) ethanamine (14i)

Yield: 73%. Brown oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.36 – 7.27 (m, 4H), 7.27 – 7.14 (m, 4H), 7.12 (d, *J* = 7.9 Hz, 2H), 3.74 (q, *J* = 6.6 Hz, 1H), 2.92 (dd, *J* = 15.3, 5.9 Hz, 2H), 2.84 – 2.76 (m, 2H), 2.51 (dd, *J* = 6.9, 4.0 Hz, 2H), 2.47 – 2.34 (m, 2H), 2.25 (s, 1H), 1.89 – 1.74 (m, 2H), 1.66 – 1.51 (m, 3H), 1.37 (d, *J* = 6.6 Hz, 3H), 1.34 – 1.16 (m, 2H). ^{13}C NMR (101 MHz, $CDCl_3$) δ143.24, 140.48, 128.89, 128.22, 128.03, 127.04, 126.73, 125.57, 61.74, 57.72, 47.75, 44.09, 42.98, 37.72, 32.18, 23.97. MS (ESI) m/z 323.2 (calcd 239.0 for $C_{22}H_{31}N_2^+$ [M + H] $^+$).

5.1.3.10 N-benzyl-2-(4-benzylpiperidin-1-yl) ethanamine (14j)

Yield: 84%. Brown oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.28 (d, *J* = 4.4 Hz, 4H), 7.26 – 7.18 (m, 3H), 7.14 (t, *J* = 7.4 Hz, 1H), 7.10 (d, *J* = 7.0 Hz, 2H), 3.75 (s, 2H), 2.79 (d, *J* = 11.5 Hz, 2H), 2.65 (t, *J* = 6.2 Hz, 2H), 2.49 (d, *J* = 7.0 Hz, 2H), 2.42 (t, *J* = 6.2 Hz, 3H), 1.82 (td, *J* = 11.7, 1.7 Hz, 2H), 1.57 (d, *J* = 12.8 Hz, 2H), 1.49 – 1.42 (m, 1H), 1.25 (qd, *J* = 12.4, 3.6 Hz, 2H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 140.35, 140.07, 128.78, 128.03, 127.83, 127.80, 126.55, 125.45, 57.78, 53.70, 53.64, 45.62, 42.91, 37.64, 31.91. MS (ESI) m/z 309.2 (calcd 309.2 for $C_{21}H_{29}N_2^+$ [M + H] $^+$).

5.1.3.11 2-(4-benzylpiperidin-1-yl)-N-phenethylethanamine (14k)

Yield: 70%. Brown oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.32 – 7.23 (m, 5H), 7.22 – 7.16 (m, 3H), 7.12 (d, *J* = 7.0 Hz, 2H), 2.90 – 2.84 (m, 2H), 2.81 (dd, *J* = 12.9, 6.8 Hz, 4H), 2.72 – 2.64 (m, 2H), 2.50 (t, *J* = 6.0 Hz, 2H), 2.46 – 2.37 (m, 2H), 2.20 (s, 1H), 1.84 (td, *J* = 11.8, 1.9 Hz, 2H), 1.58 (dd, *J* = 16.9, 6.9 Hz, 2H), 1.50 – 1.40 (m, 1H), 1.31 – 1.14 (m, 2H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 140.59, 139.94, 128.98, 128.59, 128.33, 128.01, 125.99, 125.63, 57.89, 53.89, 51.11, 46.50, 43.10, 37.80, 36.18, 32.06. MS (ESI) m/z 323.2 (calcd 323.2 for $C_{22}H_{31}N_2^+$ [M + H] $^+$).

5.1.4 General method for preparation of compounds 15-25.

Propionyl chloride (9.0mmol) was added slowly to the solution of 14a (3.0mmol) (14b-14k) and Et_3N (18.0mmol) in DCM (30mL) under ice-cooled, and the mixture was stirred at 0°C for 3h. The mixture was diluted with saturated aqueous $NaHCO_3$, and extracted with DCM, and the organic layer was washed with saturated aqueous $NaHCO_3$ and brine, and then dried over anhydrous Na_2SO_4, concentrated in vacuo. The residue was purified by column chromatography on silica gel (petroleum ether/EtOAc = 1/1) to afford the free base of compounds 15-25.

5.1.4.1 *N-(2-(4-benzylpiperidin-1-yl) ethyl)-N-phenylpropionamide (15)*

Yield: 65%. Pale yellow oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.44 – 7.34 (m, 2H), 7.31 (t, *J* = 7.3 Hz, 1H), 7.25 (dd, *J* = 10.4, 4.2 Hz, 2H), 7.21 – 7.14 (m, 3H), 7.14 – 7.07 (m, 2H), 3.88 – 3.78 (m, 2H), 2.84 (d, *J* = 11.4 Hz, 2H), 2.50 (d, *J* = 7.1 Hz, 2H), 2.48 – 2.42 (m, 2H), 2.02 (q, *J* = 7.3 Hz, 2H), 1.96 – 1.83 (m, 2H), 1.58 (d, *J* = 12.9 Hz, 2H), 1.54 – 1.42 (m, 1H), 1.29 – 1.19 (m, 2H), 1.03 (t, *J* = 7.4 Hz, 3H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 173.44, 142.68, 140.51, 129.35, 128.94, 128.28, 127.96, 127.60, 125.58, 55.62, 53.79, 46.40, 43.06, 37.68, 32.04, 27.64, 9.48. MS (ESI) m/z 351.2 (calcd 351.2 for $C_{23}H_{31}N_2O^+$ [M + H] $^+$).

5.1.4.2 N-(2-(4-benzylpiperidin-1-yl) ethyl)-N-(4-methoxyphenyl) propionamide (16)

Yield: 72%. Pale yellow oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.26 (dd, *J* = 8.0, 6.8 Hz, 2H), 7.17 (t, *J* = 7.4 Hz, 1H), 7.15 – 7.05 (m, 4H), 6.88 (t, *J* = 8.9 Hz, 2H), 3.82 (s, 3H), 3.81 – 3.76 (m, 2H), 2.85 (d, *J* = 11.3 Hz, 2H), 2.51 (d, *J* = 7.0 Hz, 2H), 2.47 – 2.42 (m, 2H), 2.01 (q, *J* = 7.5 Hz, 2H), 1.90 (t, *J* = 10.9 Hz, 2H), 1.59 (d, *J* = 12.8 Hz, 2H), 1.54 – 1.42 (m, 1H), 1.35 – 1.13 (m, 2H), 1.02 (t, *J* = 7.4 Hz, 3H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 173.97, 158.78, 140.64, 135.45, 129.39, 129.05, 128.07, 125.68, 114.53, 55.68, 55.40, 53.90, 46.47, 43.16, 37.79, 32.13, 27.66, 9.57. MS (ESI) m/z 381.2 (calcd 381.3 for $C_{24}H_{33}N_2O_2^+$ [M + H] $^+$).

5.1.4.3 N-(2-(4-benzylpiperidin-1-yl) ethyl)-N-(p-tolyl) propionamide (17)

Yield: 83%. Colorless oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.26 (t, *J* = 7.3 Hz, 2H), 7.20 – 7.11 (m, 5H), 7.05 (d, *J* = 8.2 Hz, 2H), 3.83 – 3.77 (m, 2H), 2.85 (d, *J* = 11.4 Hz, 2H), 2.50 (d, *J* = 7.1 Hz, 2H), 2.47 – 2.42 (m, 2H), 2.37 (s, 3H), 2.02 (q, *J* = 7.4 Hz, 2H), 1.90 (t, *J* = 10.8 Hz, 2H), 1.58 (d, *J* = 13.0 Hz, 2H), 1.55 – 1.42 (m, 1H), 1.32 – 1.18 (m, 2H), 1.02 (t, *J* = 7.4 Hz, 3H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 173.72, 140.63, 140.11, 137.56, 130.05,

129.04, 128.05, 125.67, 113.05, 55.69, 53.90, 46.46, 43.16, 37.78, 32.13, 27.68, 21.02, 9.58. MS (ESI) m/z 365.2 (calcd 365.3 for Chemical Formula: $C_{24}H_{33}N_2O^+$ [M + H] $^+$).

5.1.4.4 N-(2-(4-benzylpiperidin-1-yl) ethyl)-N-(4-fluorophenyl) propionamide (18)

Yield: 78%. Pale yellow oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.30 – 7.23 (m, 2H), 7.22 – 7.15 (m, 3H), 7.14 – 7.11 (m, 2H),7.10 – 7.04 (m, 2H), 3.79 (t, J = 7.1 Hz, 2H), 2.83 (d, J = 11.5 Hz, 2H), 2.51 (d, J = 7.1 Hz, 2H), 2.46 – 2.41 (m, 2H), 2.05 – 1.96 (m, 2H), 1.94 – 1.84 (m, 2H), 1.59 (d, J = 12.9 Hz, 2H), 1.55 – 1.43 (m, 1H), 1.28 – 1.16 (m, 2H), 1.03 (t, J = 7.4 Hz, 3H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 173.58, 162.91, 160.44, 140.57, 138.77, 138.74, 130.18, 130.09, 129.04, 128.06, 125.69, 116.41, 116.19, 55.68, 53.85, 46.45, 43.13, 37.77, 32.11, 27.74, 9.51. MS (ESI) m/z 369.2 (calcd 369.2 for $C_{23}H_{30}FN_2O^+$ [M + H] $^+$).

5.1.4.5 N-(2-(4-benzylpiperidin-1-yl) ethyl)-N-(3, 4, 5-trifluorophenyl) propionamide (19)

Yield: 60%. Pale yellow oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.27 (t, *J* = 7.4 Hz, 2H), 7.20 – 7.10 (m, 3H), 7.01 – 6.95 (m, 2H), 3.78 (t, *J* = 6.7 Hz, 2H), 2.89 (d, *J* = 11.4 Hz, 2H), 2.57 – 2.48 (m, 4H), 2.06 (d, *J* = 6.7 Hz, 2H), 1.99 – 1.92 (m, 2H), 1.62 (d, *J* = 12.9 Hz, 2H), 1.53 – 1.47 (m, 1H), 1.30 – 1.22 (m, 2H), 1.09 – 1.03 (m, 3H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 173.13, 152.40, 152.35, 152.30, 152.25, 149.89, 149.84, 149.79, 149.74, 140.38, 140.10, 138.16, 128.99, 128.09, 125.75, 113.54, 113.32, 55.42, 53.56, 46.20, 42.98, 37.60, 31.67, 27.73, 9.36. MS (ESI) m/z 405.2 (calcd 405.2 for $C_{23}H_{28}F_3N_2O^+$ [M + H] $^+$).

5.1.4.6 N-(2-(4-benzylpiperidin-1-yl) ethyl)-N-(pyridin-2-yl) propionamide (20)

Yield: 65%. Pale yellow oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.53 – 8.45(m, 1H), 7.73 (td, J = 7.8, 2.0 Hz, 1H), 7.29 – 7.23 (m, 3H), 7.22 – 7.14 (m, 2H), 7.13 – 7.08 (m, 2H), 3.99 – 3.93 (m, 2H), 2.82 (d, J = 11.6 Hz, 2H), 2.55 – 2.46 (m, 4H), 2.21 (q, J = 7.3 Hz, 2H), 1.90 (td, J = 11.7, 2.1 Hz, 2H), 1.56 (d, J = 13.4 Hz, 2H), 1.46 – 1.35 (m, 1H), 1.21 – 1.13 (m, 2H), 1.08 (t, J = 7.4 Hz, 3H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 173.54, 155.60, 149.02, 140.50, 138.00, 128.99, 128.00, 125.63, 122.13, 121.99, 56.26, 53.82, 45.38, 43.05, 37.62, 31.97, 27.99, 9.44. MS (ESI) m/z 352.2 (calcd 352.2 for $C_{22}H_{30}N_3O^+$ [M + H] $^+$).

5.1.4.7 N-(2-(4-benzylpiperidin-1-yl) ethyl)-N-(3-fluoropyridin-2-yl) propionamide (21)

Yield: 79%. Pale yellow oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.30 (d, J = 4.3 Hz, 1H), 7.47 – 7.39 (m, 1H), 7.30 – 7.22 (m, 3H), 7.21 – 7.14 (m, 1H), 7.12 – 7.05 (m, 2H), 3.94 (t, J = 6.8 Hz, 2H), 2.73 (d, J = 11.4 Hz, 2H), 2.52 (t, J = 6.8 Hz, 2H), 2.44 (d, J = 6.8 Hz, 2H), 2.13 (s,

2H), 1.85 (t, *J* = 10.8 Hz, 2H),1.48 (d, J = 13.0 Hz, 2H), 1.42 –1.36 (m, 1H), 1.08 (t, J = 7.4 Hz, 3H), 1.02 – 0.89 (m, 2H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 173.28, 155.72, 153.14, 144.68, 144.55, 144.29, 144.23, 140.39, 129.04, 127.97, 125.62, 124.62, 124.43, 124.03, 56.60, 53.61, 44.78, 42.98, 37.46, 31.78, 27.07, 27.05, 9.16. MS (ESI) m/z 370.2 (calcd 370.2 for $C_{22}H_{29}FN_3O^+$ [M + H] $^+$).

5.1.4.8 N-(2-(4-benzylpiperidin-1-yl) ethyl)-N-(6-(trifluoromethyl) pyridin-2-yl) propionamide (22)

Yield: 65%. Yellow oil. ^{1}H NMR (600 MHz, $CDCl_3$) δ 7.80 (t, *J* = 7.9 Hz, 1H), 7.67 (d, *J* = 6.6 Hz, 1H), 7.46 (d, *J* = 7.6 Hz, 1H), 7.24 (t, *J* = 7.5 Hz, 2H), 7.15 (t, *J* = 7.4 Hz, 1H), 7.09 (d, *J* = 7.1 Hz, 2H), 4.05 (t, *J* = 6.8 Hz, 2H), 2.80 (d, *J* = 11.5 Hz, 2H), 2.55 (t, *J* = 6.8 Hz, 2H), 2.46 (d, *J* = 7.0 Hz, 2H), 2.41 (dd, *J* = 14.5, 7.2 Hz, 2H), 1.93 – 1.88 (m, 2H), 1.53 (d, *J* = 12.7 Hz, 2H), 1.50 – 1.42 (m, 1H), 1.15 (t, *J* = 7.4 Hz, 3H), 1.09 – 1.02 (m, 2H). ^{13}C NMR (151 MHz, $CDCl_3$) δ 173.78, 155.34, 140.14, 138.58, 128.79, 127.78, 125.42, 123.54, 121.77, 119.96, 117.00, 56.55, 53.54, 44.74, 42.75, 37.28, 31.66, 28.16, 9.21. MS (ESI) m/z 420.2 (calcd 420.2 for $C_{23}H_{29}F_3N_3O^+$ [M + H] $^+$).

5.1.4.9 N-(2-(4-benzylpiperidin-1-yl) ethyl)-N-(1-phenylethyl) propionamide (23)

Yield: 63%. Pale yellow oil. Spectroscopic data of amide 23 were obtained as a mixture of two rotational isomers. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.35 – 7.28 (m, 3H), 7.25 (td, J = 7.5, 3.9 Hz, 4H), 7.21 – 7.13 (m, 1H), 7.12 – 7.06 (m, 2H), 3.23 – 3.07 (m, 2H), 2.69 – 2.60 (m, 2H), 2.47 (dd, J = 9.8, 4.9 Hz, 3H), 2.38 (dd, J = 14.8, 7.3 Hz, 2H), 2.30 – 2.12 (m, 1H), 1.96 – 1.77 (m, 2H), 1.71 (td, J = 11.7, 2.0 Hz, 1H), 1.60 (d, J = 6.9 Hz, 1H), 1.58 – 1.51 (m, 2H), 1.50 – 1.47 (m, 2H), 1.42 – 1.34 (m, 1H), 1.29 – 1.13 (m, 5H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 174.12, 173.69, 141.18, 140.75, 140.67, 140.57, 129.18, 129.16, 128.73, 128.47, 128.25, 128.21, 127.84, 127.66, 127.47, 126.91, 125.91, 125.82, 58.60, 56.75, 55.02, 54.41, 54.17, 53.79, 51.00, 43.25, 43.19, 41.49, 40.81, 37.79, 37.76, 32.08, 26.99, 26.85, 18.45, 16.80, 9.98, 9.69. MS (ESI) m/z 379.2 (calcd 379.3 for $C_{25}H_{35}N_2O^+$ [M + H] $^+$).

5.1.4.10 N-benzyl-N-(2-(4-benzylpiperidin-1-yl) ethyl) propionamide (24)

Yield: 55%. Pale yellow oil. Spectroscopic data of amide 24 were obtained as a mixture of two rotational isomers. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.33 – 7.21 (m, 6H), 7.17 – 7.09 (m, 4H), 4.61 and 4.55 (pair of s, due to rotamers, 2H), 3.53 (t, *J* = 6.6 Hz, 1H), 3.33 (s, 1H),

3.02 and 2.83 (pair of d, due to rotamers, *J* = 10.7 Hz, *J* = 10.5 Hz, 2H), 2.63 (d, *J* = 6.3 Hz, 1H), 2.50 (d, *J* = 3.2 Hz, 2H), 2.45 – 2.39 (m, 2H), 2.36 – 2.30 (m, 1H), 2.05 (t, *J* = 11.4 Hz, 1H), 1.97 – 1.89 (m, 1H), 1.61 (d, *J* = 12.2 Hz, 2H), 1.50 (s, 1H), 1.39 – 1.24 (m, 2H), 1.19 and 1.10 (pair of t, due to rotamers, *J* = 7.1 Hz, *J* = 7.3 Hz, 3H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 173.99, 173.64, 140.04, 139.96, 137.61, 136.64, 128.73, 128.52, 128.20, 127.87, 127.69, 127.18, 126.93, 125.97, 125.55, 56.24, 54.69, 53.74, 53.07, 51.16, 48.49, 43.96, 42.66, 42.61, 42.54, 37.29, 37.09, 31.42, 30.86, 28.46, 26.05, 25.84, 9.49, 9.42, 9.11. MS (ESI) m/z 365.2 (calcd 365.3 for $C_{24}H_{33}N_2O^+$ $[M + H]^+$).

5.1.4.11 N-(2-(4-benzylpiperidin-1-yl) ethyl)-N-phenethylpropionamide (25)

Yield: 51%. Pale yellow oil. Spectroscopic data of amide 25 were obtained as a mixture of two rotational isomers. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.32 – 7.24 (m, 4H), 7.23 – 7.18 (m, 2H), 7.18 – 7.11 (m, 4H), 3.54 – 3.46 (m, 3H), 3.26 – 3.21 (m, 1H), 2.91 (d, *J* = 11.6 Hz, 1H), 2.88 – 2.77 (m, 3H), 2.51 (d, *J* = 7.1 Hz, 2H), 2.50 – 2.45 (m, 1H), 2.39 – 2.30 (m, 2H), 2.14 (q, *J* = 7.4 Hz, 1H), 1.99 – 1.87 (m, 2H), 1.62 (d, *J* = 12.7 Hz, 2H), 1.55 – 1.48 (m, 1H), 1.33 – 1.20 (m, 2H), 1.15 and 1.04 (pair of t, due to rotamers, *J* = 7.4 Hz, *J* = 7.4 Hz, 3H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 173.46, 173.44, 140.48, 140.34, 139.29, 138.15, 128.96, 128.95, 128.73, 128.59, 128.31, 128.04, 128.01, 126.55, 126.11, 125.70, 125.64, 57.11, 56.00, 54.26, 54.01, 49.93, 48.62, 46.14, 43.50, 43.04, 42.96, 37.65, 37.59, 35.26, 34.01, 32.03, 31.97, 26.15, 25.88, 9.46, 9.30. MS (ESI) m/z 379.2 (calcd 379.3 for $C_{25}H_{35}N_2O^+$ $[M + H]^+$).

5.1.5 General method for preparation of 5-chloro-N-(4-methoxyphenyl) pentanamide (26c).

4-chlorobutyryl chloride (15.0 mmol) was added dropwise to a mixture of the 4-methoxyaniline (10.0 mmol) in 50 mL acetone under ice bath conditions. After the addition is completed, stirring for 4 h at room temperature, the reaction mixture was quenched with water and extracted with ethyl acetate. The organic layer was washed with water and brine successively, dried over anhydrous sodium sulfate, and concentrated in vacuum. The resulting residue was purified by silica gel chromatography (petroleum ether/EtOAc = 10/1) to give the intermediate 26c.

Yield: 67%. Pale yellow oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.01 (s, 1H), 7.39 (d, *J* = 8.9 Hz,

2H), 6.81 (d, *J* = 8.9 Hz, 2H), 3.76 (s, 3H), 3.52 (t, *J* = 5.8 Hz, 2H), 2.34 (t, *J* = 6.8 Hz, 2H), 1.83 – 1.78 (m, 4H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 171.01, 156.28, 130.92, 121.99, 113.93, 55.35, 44.52, 36.16, 31.82, 22.84. MS (ESI) m/z 242.2 (calcd 242.1 for $C_{12}H_{17}ClNO_2^+$ [M + H] $^+$).

5.1.6 General method for preparation of compounds 27a-27c.

A mixture of intermediate 26a (10mmol) (26b-26c) and 4-benzyl piperidine (11.0mmol) in 100 mL acetone was stirred at room temperature for 8 hours in the presence of potassium carbonate (30mmol). After the reaction was completed, it was quenched with water, and extracted with ethyl acetate; the mixture was washed three times with saturated brine and dried over anhydrous sodium sulfate. Distilling solvent under reduced pressure, the crude product was purified by silica gel chromatography (petroleum ether/EtOAc = 1/1) to yield corresponding intermediates 27a-27c.

5.1.6.1 3-(4-benzylpiperidin-1-yl)-N-(4-methoxyphenyl) propanamide (27a)

Yield: 90%. Brown solid, m. p. 139-141 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 10.80 (s, 1H), 7.48 – 7.42 (m, 2H), 7.28 (t, *J* = 7.4 Hz, 2H), 7.20 (t, *J* = 7.3 Hz, 1H), 7.13 (d, *J* = 7.0 Hz, 2H), 6.87 – 6.79 (m, 2H), 3.75 (s, 3H), 3.11 (d, *J* = 11.7 Hz, 2H), 2.79 (t, *J* = 6.1 Hz, 2H), 2.61 (t, *J* = 6.1 Hz, 2H), 2.56 (d, *J* = 7.0 Hz, 2H), 2.14 (t, *J* = 11.1 Hz, 2H), 1.74 (d, *J* = 12.9 Hz, 2H), 1.69 – 1.58 (m, 1H), 1.48 – 1.35 (m, 2H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 169.71, 155.68, 139.69, 131.80, 128.90, 128.09, 125.86, 120.95, 113.89, 55.25, 53.52, 52.78, 42.45, 37.10, 32.19, 31.33. MS (ESI) m/z 353.2 (calcd 353.2 for $C_{22}H_{29}N_2O_2^+$ [M + H] $^+$).

5.1.6.2 4-(4-benzylpiperidin-1-yl)-N-(4-methoxyphenyl) butanamide (27b)

Yield: 85%. Brown solid, m. p. 138-139 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 9.20 (s, 1H), 7.43 (t, *J* = 6.1 Hz, 2H), 7.30 – 7.23 (m, 2H), 7.18 (q, *J* = 6.9 Hz, 1H), 7.13 – 7.08 (m, 2H), 6.82 (t, *J* = 6.1 Hz, 2H), 3.74 (s, 3H), 2.88 (d, *J* = 11.4 Hz, 2H), 2.50 (d, *J* = 7.0 Hz, 2H), 2.40 – 2.31 (m, 4H), 1.91 – 1.80 (m, 4H), 1.67 – 1.58 (m, 2H), 1.57 – 1.50 (m, 1H), 1.33 – 1.21 (m, 2H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 171.41, 155.84, 140.24, 131.46, 128.84, 127.97, 125.62, 121.61, 113.77, 57.35, 55.20, 53.44, 42.87, 37.58, 35.34, 31.77, 22.28. MS (ESI) m/z 367.3 (calcd 367.2 for $C_{23}H_{31}N_2O_2^+$ [M + H] $^+$).

5.1.6.3 5-(4-benzylpiperidin-1-yl)-N-(4-methoxyphenyl) pentanamide (27c)

Yield: 75%. Brown solid, m. p. 116-118 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.36 (s, 1H),

7.42 (d, *J* = 9.0 Hz, 2H), 7.26 (dd, *J* = 8.4, 6.4 Hz, 2H), 7.17 (t, *J* = 7.3 Hz, 1H), 7.11 (d, *J* = 7.1 Hz, 2H), 6.80 (d, *J* = 8.9 Hz, 2H), 3.74 (s, 3H), 2.89 (d, *J* = 11.5 Hz, 2H), 2.50 (d, *J* = 7.0 Hz, 2H), 2.32 (t, *J* = 7.3 Hz, 4H), 1.86 (t, *J* = 11.1 Hz, 2H), 1.68 (dt, *J* = 14.9, 7.4 Hz, 2H), 1.61 (d, *J* = 14.7 Hz, 2H), 1.57 – 1.52 (m, 2H), 1.51 – 1.46 (m, 1H), 1.35 – 1.24 (m, 2H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 171.51, 156.05, 140.36, 131.25, 128.94, 128.03, 125.68, 121.87, 113.84, 58.05, 55.29, 53.68, 42.90, 37.61, 36.84, 31.63, 26.02, 23.58. MS (ESI) m/z 381.2 (calcd 381.2 for $C_{24}H_{33}N_2O_2^+$ $[M + H]^+$).

5.1.7 General method for preparation of compounds 28a-28c.

A mixture of intermediate 12b (10mmol) and 4-Phenylpiperidine (11.0mmol) (4-phenethyl- piperidine or 4-(3-phenylpropyl) piperidine) in 100 mL acetone was stirred at room temperature for 8 hours in the presence of potassium carbonate (30mmol). After the reaction was completed, it was quenched with water, and extracted with ethyl acetate; the mixture was washed three times with saturated brine and dried over anhydrous sodium sulfate. Distilling solvent under reduced pressure, the crude product was purified by silica gel chromatography (petroleum ether/EtOAc = 1/1) to yield corresponding intermediates 28a-28c.

5.1.7.1 N-(4-methoxyphenyl)-2-(4-phenylpiperidin-1-yl) acetamide (28a)

Yield: 72%. Brown solid, m. p. 115-117 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 9.10 (s, 1H), 7.53 – 7.48 (m, 2H), 7.32 (t, *J* = 7.5 Hz, 2H), 7.23 (dd, *J* = 15.3, 7.0 Hz, 3H), 6.90 – 6.85 (m, 2H), 3.78 (s, 3H), 3.14 (s, 2H), 3.02 (d, *J* = 11.6 Hz, 2H), 2.55 (tt, *J* = 11.9, 3.8 Hz, 1H), 2.38 (td, *J* = 11.7, 2.4 Hz, 2H), 1.91 (d, *J* = 10.7 Hz, 2H), 1.86 – 1.76 (m, 2H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 168.48, 156.32, 145.74, 130.95, 128.55, 126.81, 126.39, 121.23, 116.42, 114.21, 62.40, 55.52, 54.79, 41.81, 33.71. MS (ESI) m/z 325.2 (calcd 325.2for $C_{20}H_{25}N_2O_2^+$ $[M + H]^+$).

5.1.7.2 N-(4-methoxyphenyl)-2-(4-phenethylpiperidin-1-yl) acetamide (28b)

Yield: 84%. Brown solid, m. p. 125-126 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 9.28 (s, 1H), 7.50 (d, *J* = 8.9 Hz, 2H), 7.28 (dd, *J* = 13.7, 6.6 Hz, 2H), 7.19 (t, *J* = 7.7 Hz, 3H), 6.90 – 6.84 (m, 2H), 3.79 (s, 3H), 3.16 (s, 2H), 2.96 (d, *J* = 10.5 Hz, 2H), 2.67 – 2.60 (m, 2H), 2.31 (s, 2H), 1.81 (d, *J* = 9.5 Hz, 2H), 1.60 (dt, *J* = 7.7, 5.8 Hz, 2H), 1.41 – 1.27 (m, 3H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 168.82, 156.23, 142.35, 130.88, 128.33, 128.24, 125.73,

121.11, 116.37, 114.09, 61.96, 55.44, 54.14, 50.48, 38.10, 34.43, 32.98. MS (ESI) m/z 353.2 (calcd 353.2 for $C_{22}H_{29}N_2O_2^+$ [M + H] $^+$).

5.1.7.3 N-(4-methoxyphenyl)-2-(4-(3-phenylpropyl) piperidin-1-yl) acetamide (28c)

Yield: 87%. Brown solid, m. p. 119-122 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 9.09 (s, 1H), 7.50 – 7.44 (m, 2H), 7.29 – 7.23 (m, 2H), 7.19 – 7.13 (m, 3H), 6.87 – 6.81 (m, 2H), 3.74 (s, 3H), 3.03 (s, 2H), 2.84 (d, *J* = 11.5 Hz, 2H), 2.62 – 2.56 (m, 2H), 2.17 (t, *J* = 11.1 Hz, 2H), 1.72 – 1.66 (m, 2H), 1.61 (dd, *J* = 15.2, 7.6 Hz, 2H), 1.32 – 1.19 (m, 5H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 168.37, 155.95, 142.31, 130.76, 128.13, 128.04, 125.45, 120.82, 116.09, 113.87, 62.11, 55.42, 55.18, 54.15, 35.87, 35.83, 34.78, 32.45, 28.41. MS (ESI) m/z 367.2 (calcd 367.2 for $C_{23}H_{31}N_2O_2^+$ [M + H] $^+$).

5.1.8 General method for preparation of compounds 29a-29c, 30a-30c.

The intermediate 27a (10mmol) (27b-27c, 28a-28c) was dissolved in 100 mL of anhydrous THF and stirred for 15 min in an ice bath. Then, $LiAlH_4$ (20mmol) was added slowly, the mixture was t refluxed at 65 °C for 4 h. The reaction mixture was quenched with ethanol under 0 °C conditions, and extracted with ethyl acetate. The organic layer was washed with 5% NaOH solution, water and brine successively, and then dried over anhydrous Na_2SO_4 and concentrated in vacuum. The resulting residue was purified by silica gel chromatography (petroleum ether/EtOAc = 1/2) to give the desired product *29a-29c, 30a-30c.*

5.1.8.1 N-(3-(4-benzylpiperidin-1-yl) propyl)-4-methoxyaniline (29a)

Yield: 72%. Yellow oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.30 – 7.23 (m, 2H), 7.21 – 7.11 (m, 3H), 6.80 – 6.74 (m, 2H), 6.57 – 6.51 (m, 2H), 3.89 (s, 1H), 3.72 (s, 3H), 3.09 (t, *J* = 6.4 Hz, 2H), 2.91 (d, *J* = 11.6 Hz, 2H), 2.54 (d, *J* = 7.0 Hz, 2H), 2.41 (t, *J* = 6.8 Hz, 2H), 1.88 – 1.80 (m, 2H), 1.80 – 1.71 (m, 2H), 1.64 (d, *J* = 13.1 Hz, 2H), 1.54 – 1.46 (m, 1H), 1.38 – 1.24 (m, 2H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 151.67, 142.97, 140.44, 128.97, 128.93, 127.99, 125.62, 114.73, 113.79, 57.43, 55.63, 53.84, 44.32, 43.01, 37.77, 32.12, 25.99. MS (ESI) m/z 339.2 (calcd 339.2 for $C_{22}H_{31}N_2O^+$ [M + H] $^+$).

5.1.8.2 N-(4-(4-benzylpiperidin-1-yl) butyl)-4-methoxyaniline (29b)

Yield: 67%. Yellow oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.31 – 7.24 (m, 3H), 7.21 – 7.11 (m, 3H), 6.81 – 6.74 (m, 2H), 6.59 – 6.54 (m, 1H), 3.91 (s, 1H), 3.74 (s, 3H), 3.07 (t, *J* = 6.3 Hz,

2H), 2.94 – 2.80 (m, 2H), 2.52 (t, *J* = 5.5 Hz, 2H), 2.29 (dt, *J* = 15.2, 7.4 Hz, 2H), 1.89 – 1.74 (m, 2H), 1.63 – 1.58 (m, 4H), 1.56 – 1.45 (m, 3H), 1.38 – 1.27 (m, 2H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 151.86, 142.75, 140.67, 129.07, 128.11, 125.72, 114.85, 113.92, 58.54, 55.79, 53.89, 44.77, 43.18, 37.94, 32.12, 27.55, 24.76. MS (ESI) m/z 353.2 (calcd 353.3 for $C_{23}H_{33}N_2O^+$ [M + H] $^+$).

5.1.8.3 N-(5-(4-benzylpiperidin-1-yl) pentyl)-4-methoxyaniline (29c)

Yield: 62%. Yellow oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.27 (dd, *J* = 10.1, 4.6 Hz, 2H), 7.21 – 7.12 (m, 4H), 6.80 – 6.74 (m, 2H), 6.59 – 6.53 (m, 1H), 3.85 (s, 1H), 3.74 (s, 3H), 3.05 (t, *J* = 7.1 Hz, 2H), 2.88 (t, *J* = 12.1 Hz, 2H), 2.52 (dd, *J* = 6.9, 3.2 Hz, 2H), 2.33 – 2.23 (m, 2H), 1.82 (dd, *J* = 21.3, 11.2 Hz, 2H), 1.62 (dd, *J* = 14.4, 7.1 Hz, 4H), 1.55 – 1.44 (m, 3H), 1.38 (dt, *J* = 11.7, 5.7 Hz, 2H), 1.33 – 1.22 (m, 2H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 151.93, 142.74, 140.69, 129.08, 128.10, 125.71, 114.85, 113.98, 58.95, 55.79, 53.96, 44.86, 43.18, 37.94, 32.11, 29.54, 26.84, 25.23. MS (ESI) m/z 367.2 (calcd 367.3 for $C_{24}H_{35}N_2O^+$ [M + H] $^+$).

5.1.8.4 4-methoxy-N-(2-(4-phenylpiperidin-1-yl) ethyl) aniline (30a)

Yield: 75%. Brown oil. ^{1}H NMR (600 MHz, $CDCl_3$) δ 7.31 – 7.27 (m, 2H), 7.24 – 7.17 (m, 3H), 6.81 – 6.77 (m, 2H), 6.63 – 6.60 (m, 2H), 3.73 (s, 1H), 3.71 (s, 3H), 3.14 (t, *J* = 6.0 Hz, 2H), 3.02 (d, *J* = 11.5 Hz, 2H), 2.63 (t, *J* = 6.0 Hz, 2H), 2.50 (tt, *J* = 11.8, 4.1 Hz, 1H), 2.09 (td, *J* = 11.6, 2.6 Hz, 2H), 1.82 (d, *J* = 12.2 Hz, 2H), 1.80 – 1.74 (m, 2H). ^{13}C NMR (151 MHz, $CDCl_3$) δ 151.96, 146.21, 142.87, 128.30, 126.72, 126.03, 114.80, 114.12, 57.15, 55.71, 54.10, 42.55, 41.52, 33.38. MS (ESI) m/z 311.2 (calcd 311.2 for $C_{20}H_{27}N_2O^+$ [M + H] $^+$).

5.1.8.5 4-methoxy-N-(2-(4-phenethylpiperidin-1-yl) ethyl) aniline (30b)

Yield: 69%. Brown oil. ^{1}H NMR (600 MHz, $CDCl_3$) δ 7.30 – 7.25 (m, 2H), 7.17 (dt, *J* = 9.0, 3.8 Hz, 3H), 6.80 – 6.76 (m, 2H), 6.62 – 6.59 (m, 2H), 3.75 (s,1H), 3.74 (s, 3H), 3.11 (t, *J* = 6.0 Hz, 2H), 2.89 (d, *J* = 11.0 Hz, 2H), 2.62 (dd, *J* = 9.1, 7.0 Hz, 2H), 2.58 (dd, *J* = 11.3, 5.2 Hz, 2H), 1.95 (t, *J* = 10.4 Hz, 2H), 1.72 (d, *J* = 9.1 Hz, 2H), 1.59 – 1.54 (m, 2H), 1.30 – 1.25 (m, 3H). ^{13}C NMR (151 MHz, $CDCl_3$) δ 152.01, 142.98, 142.73, 128.28, 128.27, 125.60, 114.86, 114.18, 57.21, 55.82, 53.75, 41.58, 38.37, 35.37, 33.08, 32.31. MS (ESI) m/z 339.2 (calcd 339.2 for $C_{22}H_{31}N_2O^+$ [M + H] $^+$).

5.1.8.6 4-methoxy-N-(2-(4-(3-phenylpropyl) piperidin-1-yl) ethyl) aniline (30c)

Yield: 82%. Brown oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.24 (dd, *J* = 9.3, 5.5 Hz, 2H), 7.17 – 7.11 (m, 3H), 6.77 – 6.74 (m, 2H), 6.59 – 6.54 (m, 2H), 3.87 (s, 1H), 3.69 (s, 3H), 3.07 (t, *J* = 6.0 Hz, 2H), 2.84 (d, *J* = 11.3 Hz, 2H), 2.54 (dt, *J* = 12.3, 6.8 Hz, 4H), 1.90 (t, *J* = 11.0 Hz, 2H), 1.63 (d, *J* = 10.6 Hz, 2H), 1.61 – 1.55 (m, 1H), 1.29 – 1.15 (m, 6H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 151.76, 142.79, 142.44, 128.14, 128.03, 125.42, 116.12, 114.63, 113.94, 57.05, 55.53, 53.66, 41.37, 36.02, 35.95, 35.54, 32.20, 28.50. MS (ESI) m/z 353.2 (calcd 353.3 for $C_{23}H_{33}N_2O^+$ $[M + H]^+$).

5.1.9 General method for preparation of compounds 31-36.

Propionyl chloride (9.0mmol) was added slowly to the solution of 29a (3.0mmol) (29b-29c, 30a-30c) and Et_3N (18.0mmol) in DCM (30mL) under ice-cooled, and the mixture was stirred at 0°C for 3h. The mixture was diluted with saturated aqueous $NaHCO_3$, and extracted with DCM, and the organic layer was washed with saturated aqueous $NaHCO_3$ and brine, and then dried over anhydrous Na_2SO_4, concentrated in vacuo. The residue was purified by column chromatography on silica gel to (petroleum ether/EtOAc = 1/1) afford the free base of compounds 31-36.

5.1.9.1 N-(3-(4-benzylpiperidin-1-yl) propyl)-N-(4-methoxyphenyl) propionamide (31)

Yield: 58%. Yellow oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.25 (t, *J* = 7.3 Hz, 2H), 7.16 (t, *J* = 7.3 Hz, 2H), 7.13 – 7.05 (m, 3H), 6.91 (d, *J* = 8.8 Hz, 2H), 3.80 (s, 3H), 3.67 (t, *J* = 7.1 Hz, 2H), 3.16 (d, *J* = 11.6 Hz, 2H), 2.62 – 2.56 (m, 2H), 2.52 (d, *J* = 6.7 Hz, 2H), 2.14 (t, *J* = 11.1 Hz, 2H), 2.02 (q, *J* = 7.4 Hz, 2H), 1.82 (dt, *J* = 14.9, 7.4 Hz, 2H), 1.66 (d, *J* = 12.9 Hz, 2H), 1.60 – 1.54 (m, 1H), 1.51 – 1.45 (m, 2H), 1.01 (t, *J* = 7.4 Hz, 3H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 173.84, 158.57, 139.60, 134.47, 128.81, 128.58, 127.79, 125.51, 114.43, 54.98, 54.68, 52.50, 46.70, 42.20, 36.70, 34.92, 30.03, 28.86, 27.23, 23.54, 9.16. MS (ESI) m/z 395.2 (calcd 395.3 for $C_{25}H_{35}N_2O_2^+$ $[M + H]^+$).

5.1.9.2 N-(4-(4-benzylpiperidin-1-yl) butyl)-N-(4-methoxyphenyl) propionamide (32)

Yield: 52%. Beige oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.27 (dd, *J* = 12.2, 5.0 Hz, 2H), 7.20 – 7.11 (m, 3H), 7.05 (d, *J* = 8.8 Hz, 2H), 6.91 (d, *J* = 8.8 Hz, 2H), 3.82 (s, 3H), 3.65 (t, *J* = 6.9 Hz, 2H), 2.94 (d, *J* = 11.4 Hz, 2H), 2.52 (d, *J* = 7.0 Hz, 2H), 2.39 – 2.33 (m, 2H), 2.01 (q, *J* = 7.5 Hz, 2H), 1.91 (t, *J* = 11.2 Hz, 2H), 1.63 (d, *J* = 12.5 Hz, 2H), 1.55 – 1.46 (m, 5H),

1.43 – 1.31 (m, 2H), 1.01 (t, *J* = 7.5 Hz, 3H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 173.92, 158.76, 140.39, 135.11, 129.15, 128.94, 128.03, 125.66, 114.62, 58.27, 55.32, 53.58, 48.64, 42.89, 37.60, 31.51, 29.54, 27.60, 25.57, 23.55, 9.53. MS (ESI) m/z 409.2 (calcd 409.3 for $C_{26}H_{37}N_2O_2^+$ [M + H] $^+$).

5.1.9.3 N-(5-(4-benzylpiperidin-1-yl) pentyl)-N-(4-methoxyphenyl) propionamide (33)

Yield: 61%. Beige oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.26 (t, *J* = 7.8 Hz, 2H), 7.18 (d, *J* = 7.2 Hz, 1H), 7.13 (t, *J* = 6.6 Hz, 2H), 7.04 (d, *J* = 8.8 Hz, 2H), 6.91 (d, *J* = 8.8 Hz, 2H), 3.82 (s, 3H), 3.66 – 3.61 (m, 2H), 2.88 (d, *J* = 11.5 Hz, 2H), 2.52 (d, *J* = 7.0 Hz, 2H), 2.28 – 2.23 (m, 2H), 2.01 (q, *J* = 7.4 Hz, 2H), 1.83 (t, *J* = 10.9 Hz, 2H), 1.61 (d, *J* = 12.6 Hz, 2H), 1.53 – 1.45 (m, 5H), 1.36 – 1.25 (m, 4H), 1.02 (t, *J* = 7.5 Hz, 3H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 173.83, 158.73, 140.57, 135.28, 129.18, 128.98, 128.01, 125.62, 114.60, 58.69, 55.33, 53.71, 48.97, 43.05, 37.80, 31.88, 27.64, 27.51, 26.48, 24.74, 9.57. MS (ESI) m/z 423.2 (calcd 423.3 for $C_{27}H_{39}N_2O_2^+$ [M + H] $^+$).

5.1.9.4 N-(4-methoxyphenyl)-N-(2-(4-phenylpiperidin-1-yl) ethyl) propionamide (34)

Yield: 43%. Colorless oil. ^{1}H NMR (600 MHz, $CDCl_3$) δ 7.28 (dd, *J* = 10.0, 5.2 Hz, 2H), 7.20 (d, *J* = 7.1 Hz, 2H), 7.17 (dd, *J* = 10.2, 4.3 Hz, 1H), 7.15 – 7.11 (m, 2H), 6.93 – 6.89 (m, 2H), 3.86 – 3.83 (m, 2H), 3.81 (s, 3H), 3.00 (d, *J* = 11.5 Hz, 2H), 2.54 – 2.51 (m, 2H), 2.46 (tt, *J* = 11.8, 3.8 Hz, 1H), 2.09 (td, *J* = 11.7, 2.3 Hz, 2H), 2.04 (q, *J* = 7.5 Hz, 2H), 1.79 (d, *J* = 10.9 Hz, 2H), 1.72 (qd, *J* = 12.5, 3.6 Hz, 2H), 1.04 (t, *J* = 7.5 Hz, 3H). ^{13}C NMR (151 MHz, $CDCl_3$) δ 173.85, 158.69, 146.19, 135.34, 129.25, 128.16, 126.61, 125.86, 114.44, 55.66, 55.25, 54.19, 46.35, 42.35, 33.27, 27.54, 9.48. MS (ESI) m/z 367.2 (calcd 367.2 for $C_{23}H_{31}N_2O_2^+$ [M + H] $^+$).

5.1.9.5 N-(4-methoxyphenyl)-N-(2-(4-phenethylpiperidin-1-yl) ethyl) propionamide (35)

Yield: 49%. Colorless oil. ^{1}H NMR (600 MHz, $CDCl_3$) δ 7.25 (dd, *J* = 8.9, 5.9 Hz, 2H), 7.18 – 7.14 (m, 3H), 7.12 – 7.08 (m, 2H), 6.91 – 6.88 (m, 2H), 3.81 (s, 3H), 3.79 (d, *J* = 7.3 Hz, 2H), 2.87 (d, *J* = 11.5 Hz, 2H), 2.60 (dd, *J* = 9.1, 7.1 Hz, 2H), 2.47 – 2.43 (m, 2H), 2.02 (q, *J* = 7.5 Hz, 2H), 1.93 (t, *J* = 10.9 Hz, 2H), 1.68 (d, *J* = 9.6 Hz, 2H), 1.56 – 1.51 (m, 2H), 1.27 – 1.17 (m, 3H), 1.02 (t, *J* = 7.5 Hz, 3H). ^{13}C NMR (151 MHz, $CDCl_3$) δ 173.88, 158.73, 142.66, 135.43, 129.32, 128.17, 128.14, 125.47, 114.47, 55.73, 55.32, 53.87, 46.44, 38.35, 35.19, 32.98, 32.24, 27.60, 9.53. MS (ESI) m/z 395.2 (calcd 395.3 for $C_{25}H_{35}N_2O_2^+$

$[M + H]^+$).

5.1.9.6 N-(4-methoxyphenyl)-N-(2-(4-(3-phenylpropyl) piperidin-1-yl) ethyl) propionamide (36)

Yield: 66%. Beige oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.28 – 7.22 (m, 2H), 7.15 (dd, *J* = 6.9, 4.0 Hz, 3H), 7.08 (d, *J* = 8.8 Hz, 2H), 6.89 (d, *J* = 8.8 Hz, 2H), 3.87 – 3.81 (m, 2H), 3.80 (s, 3H), 3.00 (d, *J* = 10.6 Hz, 2H), 2.62 – 2.54 (m, 4H), 2.10 – 1.98 (m, 4H), 1.65 (d, *J* = 8.7 Hz, 2H), 1.61 – 1.53 (m, 1H), 1.31 – 1.19 (m, 6H), 1.05 – 0.98 (m, 3H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 173.89, 158.66, 142.27, 135.01, 128.99, 128.04, 127.94, 125.34, 114.43, 55.13, 54.83, 53.24, 45.73, 35.81, 35.68, 34.96, 31.24, 28.46, 28.35, 27.37, 9.31. MS (ESI) m/z 409.2 (calcd 409.3 for $C_{26}H_{37}N_2O_2^+$ $[M + H]^+$).

5.1.10 General method for preparation of compounds 37a-37b, 38a-38b, 39a-39b.

A mixture of intermediate 12b (10mmol)(12c, 12d) and 4-(4-methylbenzyl) piperidine or (11.0 mmol) (4-(4-fluorobenzyl) piperidine) in 100 mL acetone was stirred at room temperature for 8 hours in the presence of potassium carbonate (30mmol). After the reaction was completed, it was quenched with water, and extracted with ethyl acetate; the mixture was washed three times with saturated brine and dried over anhydrous sodium sulfate. Distilling solvent under reduced pressure, the crude product was purified by silica gel chromatography (petroleum ether/EtOAc = 1/1) to yield corresponding intermediates 37a-37b, 38a-38b, and 39a-39b.

5.1.10.1 N-(4-methoxyphenyl)-2-(4-(4-methylbenzyl) piperidin-1-yl) acetamide (37a)

Yield: 92%. Beige solid, m. p. 120-121 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 9.08 (s, 1H), 7.48 (d, *J* = 8.9 Hz, 2H), 7.07 (d, *J* = 7.9 Hz, 2H), 7.01 (d, *J* = 7.9 Hz, 2H), 6.84 (d, *J* = 8.9 Hz, 2H), 3.73 (s, 3H), 3.03 (s, 2H), 2.83 (d, *J* = 11.5 Hz, 2H), 2.49 (d, *J* = 7.1 Hz, 2H), 2.30 (s, 3H), 2.13 (t, *J* = 11.0 Hz, 2H), 1.65 (d, *J* = 12.4 Hz, 2H), 1.56 – 1.48 (m, 1H), 1.30 (qd, *J* = 12.5, 3.6 Hz, 2H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 168.23, 155.90, 136.92, 134.98, 130.72, 128.67, 128.64, 120.79, 113.82, 62.01, 55.11, 54.00, 42.31, 37.01, 32.19, 20.75. MS (ESI) m/z 353.2 (calcd 353.2 for $C_{22}H_{29}N_2O_2^+$ $[M + H]^+$).

5.1.10.2 2-(4-(4-fluorobenzyl) piperidin-1-yl)-N-(4-methoxyphenyl) acetamide (37b)

Yield: 90%. Beige solid, m. p. 133-135 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 9.07 (s, 1H), 7.51 – 7.44 (m, 2H), 7.13 – 7.06 (m, 2H), 7.01 – 6.93 (m, 2H), 6.91 – 6.84 (m, 2H), 3.79 (s,

3H), 3.07 (s, 2H), 2.88 (d, *J* = 11.7 Hz, 2H), 2.54 (d, *J* = 7.1 Hz, 2H), 2.19 (td, *J* = 11.7, 2.0 Hz, 2H), 1.67 (d, *J* = 13.0 Hz, 2H), 1.59 – 1.46 (m, 1H), 1.37 – 1.22 (m, 2H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 168.47, 162.50, 160.08, 156.20, 135.85, 135.82, 130.86, 130.33, 130.25, 121.07, 115.06, 114.85, 114.10, 62.21, 55.44, 54.21, 42.13, 37.28, 32.35. MS (ESI) m/z 357.2 (calcd 357.2 for $C_{21}H_{26}FN_2O_2^+$ [M + H] $^+$).

5.1.10.3 2-(4-(4-methylbenzyl) piperidin-1-yl)-N-(p-tolyl) acetamide (38a)

Yield: 88%. White solid, m. p. 126-128 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 9.14 (s, 1H), 7.45 (d, *J* = 8.3 Hz, 2H), 7.11 (dd, *J* = 12.9, 8.1 Hz, 4H), 7.03 (d, *J* = 7.9 Hz, 2H), 3.05 (s, 2H), 2.86 (d, *J* = 11.6 Hz, 2H), 2.52 (d, *J* = 7.1 Hz, 2H), 2.31 (d, *J* = 3.0 Hz, 6H), 2.16 (td, *J* = 11.7, 2.0 Hz, 2H), 1.68 (d, *J* = 12.8 Hz, 2H), 1.59 – 1.46 (m, 1H), 1.37 – 1.24 (m, 2H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 168.59, 137.11, 135.25, 135.08, 133.51, 129.36, 128.84, 128.83, 119.34, 62.26, 54.20, 42.50, 37.22, 32.42, 20.91, 20.77. MS (ESI) m/z 337.2 (calcd 337.2 for $C_{22}H_{29}N_2O^+$ [M + H] $^+$).

5.1.10.4 2-(4-(4-fluorobenzyl) piperidin-1-yl)-N-(p-tolyl) acetamide (38b)

Yield: 94%. Beige solid, m. p. 119-121 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 9.16 (s, 1H), 7.45 (d, *J* = 8.4 Hz, 2H), 7.15 – 7.07 (m, 4H), 7.00 – 6.93 (m, 2H), 3.08 (s, 2H), 2.89 (d, *J* = 11.7 Hz, 2H), 2.54 (d, *J* = 7.1 Hz, 2H), 2.31 (s, 3H), 2.25 – 2.16 (m, 2H), 1.67 (d, *J* = 13.1 Hz, 2H), 1.58 – 1.46 (m, 1H), 1.38 – 1.24 (m, 2H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 168.48, 162.49, 160.07, 135.81, 135.78, 135.04, 133.67, 130.31, 130.24, 129.42, 119.43, 115.05, 114.84, 62.19, 54.14, 42.08, 37.23, 32.26, 20.81. MS (ESI) m/z 341.2 (calcd 341.2 for $C_{21}H_{26}FN_2O^+$ [M + H] $^+$).

5.1.10.5 N-(4-fluorophenyl)-2-(4-(4-methylbenzyl) piperidin-1-yl) acetamide (39a)

Yield: 89%. White solid, m. p. 130-132 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 9.21 (s, 1H), 7.56 – 7.51 (m, 2H), 7.08 (d, *J* = 7.8 Hz, 2H), 7.04 – 6.95 (m, 4H), 3.03 (s, 2H), 2.83 (d, *J* = 11.6 Hz, 2H), 2.50 (d, *J* = 7.1 Hz, 2H), 2.30 (s, 3H), 2.14 (dd, *J* = 11.5, 10.2 Hz, 2H), 1.66 (d, *J* = 12.2 Hz, 2H), 1.60 – 1.44 (m, 1H), 1.36 – 1.26 (m, 2H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 168.53, 160.09, 157.67, 136.89, 135.04, 133.62, 128.68, 120.86, 120.78, 115.38, 115.16, 61.98, 54.02, 42.31, 37.00, 32.20, 20.75. MS (ESI) m/z 341.2 (calcd 341.2 for $C_{21}H_{26}FN_2O^+$ [M + H] $^+$).

5.1.10.6 2-(4-(4-fluorobenzyl) piperidin-1-yl)-N-(4-fluorophenyl) acetamide (39b)

Yield: 83%. White solid, m. p. 127-129 °C. ^{1}H NMR (400 MHz, $CDCl_3$) δ 9.24 (s,1H), 7.57 – 7.51 (m, 2H), 7.11 – 7.05 (m, 2H), 7.05 – 6.93 (m, 4H), 3.08 (s, 2H), 2.87 (d, *J* = 11.7 Hz, 2H), 2.52 (d, *J* = 7.1 Hz, 2H), 2.19 (td, *J* = 11.7, 1.9 Hz, 2H), 1.66 (d, *J* = 12.7 Hz, 2H), 1.58 – 1.46 (m, 1H), 1.39 – 1.27 (m, 2H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 168.55, 162.33, 160.21, 159.91, 157.79, 135.69, 135.66, 133.61, 133.58, 130.20, 130.13, 121.01, 120.93, 115.48, 115.26, 114.89, 114.68, 61.95, 54.00, 41.90, 37.03, 32.09. MS (ESI) m/z 345.2 (calcd 345.2 for $C_{20}H_{23}FN_2O^+$ $[M + H]^+$).

5.1.11 General method for preparation of compounds 40a-40b, 41a-41b, 42a-42b.

The intermediate 37a (10mmol) (37b, 38a-38b, 39a-39b) was dissolved in 100 mL of anhydrous THF and stirred for 15 min in an ice bath. Then, $LiAlH_4$ (20mmol) was added slowly, the mixture was t refluxed at 65 °C for 4 h. The reaction mixture was quenched with ethanol under 0 °C conditions, and extracted with ethyl acetate. The organic layer was washed with 5% NaOH solution, water and brine successively, and then dried over anhydrous Na_2SO_4 and concentrated in vacuum. The resulting residue was purified by silica gel chromatography (petroleum ether/EtOAc = 1/2) to give the desired product 40a-40b, 41a-41b, and 42a-42b.

5.1.11.1 4-methoxy-N-(2-(4-(4-methylbenzyl) piperidin-1-yl) ethyl) aniline (40a)

Yield: 77%. Yellow oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.06 (d, *J* = 7.9 Hz, 2H), 7.00 (d, *J* = 8.0 Hz, 2H), 6.79 – 6.73 (m, 2H), 6.60 – 6.53 (m, 2H), 3.86 (s, 1H), 3.69 (s, 3H), 3.05 (t, *J* = 6.0 Hz, 2H), 2.83 (d, *J* = 11.5 Hz, 2H), 2.51 (t, *J* = 6.0 Hz, 2H), 2.46 (d, *J* = 7.0 Hz, 2H), 2.29 (s, 3H), 1.85 (dd, *J* = 16.4, 6.6 Hz, 2H), 1.59 (d, *J* = 12.5 Hz, 2H), 1.52 – 1.45 (m, 1H), 1.25 (qd, *J* = 12.4, 3.6 Hz, 2H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 151.78, 142.78, 137.34, 134.88, 128.77, 128.65, 114.64, 113.93, 57.00, 55.51, 53.56, 42.55, 41.37, 37.84, 32.03, 20.82. MS (ESI) m/z 339.2 (calcd 339.2 for $C_{22}H_{31}N_2O^+$ $[M + H]^+$).

5.1.11.2 N-(2-(4-(4-fluorobenzyl) piperidin-1-yl) ethyl)-4-methoxyaniline (40b)

Yield: 65%. Yellow oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.13 – 7.04 (m, 2H), 7.00 – 6.91 (m, 2H), 6.83 – 6.74 (m, 2H), 6.65 – 6.57 (m, 2H), 3.82 (s, 1H), 3.75 (s, 3H), 3.11 (t, *J* = 6.0 Hz, 2H), 2.89 (d, *J* = 11.5 Hz, 2H), 2.58 (t, *J* = 6.0 Hz, 2H), 2.50 (d, *J* = 7.1 Hz, 2H), 1.91 (dd, *J* = 16.6, 6.7 Hz, 2H), 1.60 (d, *J* = 12.7 Hz, 2H), 1.55 – 1.45 (m, 1H), 1.32 – 1.23 (m, 2H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 162.47, 160.05, 152.03, 142.93, 136.22, 136.18, 130.37,

130.29, 114.98, 114.86, 114.77, 114.19, 57.13, 55.83, 53.68, 42.29, 41.55, 38.03, 32.06. MS (ESI) m/z 343.2 (calcd 343.2 for $C_{21}H_{28}FN_2O^+$ $[M + H]^+$).

5.1.11.3 4-methyl-N-(2-(4-(4-methylbenzyl) piperidin-1-yl) ethyl) aniline (41a)

Yield: 69%. Brown oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.30 (d, *J* = 7.8 Hz, 2H), 7.22 (dd, *J* = 14.2, 8.0 Hz, 4H), 6.77 (d, *J* = 8.2 Hz, 2H), 4.35 (s, 1H), 3.31 (t, *J* = 6.0 Hz, 2H), 3.07 (d, *J* = 11.3 Hz, 2H), 2.75 (t, *J* = 6.0 Hz, 2H), 2.70 (d, *J* = 7.0 Hz, 2H), 2.53 (s, 3H), 2.46 (s, 3H), 2.09 (t, *J* = 11.0 Hz, 2H), 1.83 (d, *J* = 12.6 Hz, 2H), 1.77 – 1.65 (m, 1H), 1.55 – 1.44 (m, 2H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 146.24, 137.33, 134.86, 129.46, 128.77, 128.65, 125.97, 112.87, 56.91, 53.52, 42.57, 40.75, 37.86, 32.04, 20.83, 20.23. MS (ESI) m/z 323.2 (calcd 323.2 for $C_{22}H_{31}N_2^+$ $[M + H]^+$).

5.1.11.4 N-(2-(4-(4-fluorobenzyl) piperidin-1-yl) ethyl)-4-methylaniline (41b)

Yield: 75%. Brown oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.11 – 7.04 (m, 2H), 7.00 – 6.91 (m, 4H), 6.56 (d, *J* = 8.4 Hz, 2H), 4.06 (s, 1H), 3.12 (t, *J* = 6.0 Hz, 2H), 2.87 (d, *J* = 11.5 Hz, 2H), 2.56 (t, *J* = 6.1 Hz, 2H), 2.50 (d, *J* = 7.1 Hz, 2H), 2.23 (s, 3H), 1.90 (td, *J* = 11.7, 1.9 Hz, 2H), 1.59 (d, *J* = 12.6 Hz, 2H), 1.54 – 1.39 (m, 1H), 1.31 – 1.20 (m, 2H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 162.53, 160.12, 146.48, 136.32, 136.29, 130.44, 130.36, 129.73, 129.16, 128.21, 126.49, 125.83, 115.05, 114.84, 113.19, 57.16, 53.72, 42.38, 41.06, 38.13, 32.18, 20.43. MS (ESI) m/z 327.2 (calcd 327.2 for $C_{21}H_{28}FN_2^+$ $[M + H]^+$).

5.1.11.5 4-fluoro-N-(2-(4-(4-methylbenzyl) piperidin-1-yl) ethyl) aniline (42a)

Yield: 64%. Brown oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.08 (d, *J* = 7.9 Hz, 2H), 7.02 (d, *J* = 8.0 Hz, 2H), 6.91 – 6.83 (m, 2H), 6.57 – 6.51 (m, 2H), 4.19 (s, 1H), 3.07 (t, *J* = 6.0 Hz, 2H), 2.85 (d, *J* = 11.5 Hz, 2H), 2.54 (t, *J* = 6.0 Hz, 2H), 2.49 (d, *J* = 7.1 Hz, 2H), 2.31 (s, 3H), 1.89 (td, *J* = 11.7, 1.9 Hz, 2H), 1.62 (d, *J* = 12.9 Hz, 2H), 1.54 – 1.45 (m, 1H), 1.33 – 1.22 (m, 2H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 156.81, 154.48, 145.00, 144.98, 137.48, 135.13, 128.92, 128.79, 115.58, 115.36, 113.61, 113.53, 56.93, 53.66, 42.68, 41.16, 37.98, 32.16, 20.94. MS (ESI) m/z 327.2 (calcd 327.2 for $C_{21}H_{28}FN_2^+$ $[M + H]^+$).

5.1.11.6 4-fluoro-N-(2-(4-(4-fluorobenzyl) piperidin-1-yl) ethyl) aniline (42b)

Yield: 66%. Brown oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.11 – 7.05 (m, 2H), 6.98 – 6.92 (m, 2H), 6.91 – 6.86 (m, 2H), 6.58 – 6.52 (m, 2H), 4.15 (s, 1H), 3.09 (t, *J* = 6.0 Hz, 2H), 2.87 (d, *J* = 11.6 Hz, 2H), 2.56 (t, *J* = 6.0 Hz, 2H), 2.50 (d, *J* = 7.1 Hz, 2H), 1.91 (td, *J* = 11.8, 2.0

Hz, 2H), 1.60 (d, *J* = 12.7 Hz, 2H), 1.54 – 1.45 (m, 1H), 1.26 (qd, *J* = 12.3, 3.7 Hz, 2H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 162.45, 160.03, 156.86, 154.53, 145.00, 144.99, 136.19, 136.15, 130.35, 130.28, 115.63, 115.41, 114.96, 114.76, 113.64, 113.57, 56.93, 53.62, 42.27, 41.17, 38.01, 32.06. MS (ESI) m/z 331.2 (calcd 331.2 for $C_{22}H_{25}F_2N_2^+$ [M + H] $^+$).

5.1.12 General method for preparation of compounds 43-48.

Propionyl chloride (9.0mmol) was added slowly to the solution of 40a (3.0mmol) (40b, 41a-41c, 42a-42b) and Et_3N (18.0mmol) in DCM (30mL) under ice-cooled, and the mixture was stirred at 0°C for 3h. The mixture was diluted with saturated aqueous $NaHCO_3$, and extracted with DCM, and the organic layer was washed with saturated aqueous $NaHCO_3$ and brine, and then dried over anhydrous Na_2SO_4, concentrated in vacuo. The residue was purified by column chromatography on silica gel (petroleum ether/EtOAc = 1/1) to afford the free base of compounds 43-48.

5.1.12.1 N-(4-methoxyphenyl)-N-(2-(4-(4-methylbenzyl) piperidin-1-yl) ethyl) propionamide (43)

Yield: 57%. Colorless oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.18 (d, *J* = 8.1 Hz, 2H), 7.06 (dd, *J* = 8.0, 6.2 Hz, 4H), 7.01 (d, *J* = 8.0 Hz, 2H), 3.84 – 3.76 (m, 2H), 2.84 (d, *J* = 11.5 Hz, 2H), 2.45 (t, *J* = 8.2 Hz, 4H), 2.36 (s, 3H), 2.30 (s, 3H), 2.02 (q, *J* = 7.4 Hz, 2H), 1.89 (td, *J* = 11.6, 1.8 Hz, 2H), 1.58 (d, *J* = 12.6 Hz, 2H), 1.52 – 1.38 (m, 1H), 1.29 – 1.16 (m, 2H), 1.02 (t, *J* = 7.5 Hz, 3H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 173.66, 140.07, 137.50, 137.47, 134.98, 130.00, 128.85, 128.68, 128.00, 55.63, 53.84, 46.42, 42.64, 37.78, 32.06, 27.61, 20.95, 20.87, 9.52. MS (ESI) m/z 395.2 (calcd 395.3 for $C_{25}H_{35}N_2O_2^+$ [M + H] $^+$).

5.1.12.2 N-(2-(4-(4-fluorobenzyl) piperidin-1-yl) ethyl)-N-(4-methoxyphenyl) propionamide (44)

Yield: 61%. Colorless oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.14 – 7.03 (m, 4H), 7.01 – 6.86 (m, 4H), 3.83 (s, 3H), 3.82 – 3.76 (m, 2H), 2.86 (d, J = 11.3 Hz, 2H), 2.46 (dd, J = 15.6, 7.2 Hz, 4H), 2.01 (q, J = 7.5 Hz, 2H), 1.90 (t, J = 11.0 Hz, 2H), 1.57 (d, J = 12.8 Hz, 2H), 1.45 – 1.37 (m,1H), 1.30 – 1.14 (m, 2H), 1.02 (t, J = 7.5 Hz, 3H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 173.99, 162.40, 159.98, 158.79, 136.21, 136.18, 135.44, 130.34, 130.27, 129.39, 114.90, 114.69, 114.54, 55.67, 55.41, 53.86, 46.46, 42.28, 37.86, 32.03, 27.67, 9.59. HRMS (ESI) m/z 399.2431 (calcd 399.2442 for $C_{24}H_{32}FN_2O_2^+$ [M + H] $^+$).

5.1.12.3 N-(2-(4-(4-methylbenzyl) piperidin-1-yl) ethyl)-N-(p-tolyl) propionamide (45)

Yield: 57%. Colorless oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.12 – 7.05 (m, 4H), 7.01 (d, *J* = 7.9 Hz, 2H), 6.92 – 6.87 (m, 2H), 3.82 (s, 3H), 3.81 – 3.76 (m, 2H), 2.85 (d, *J* = 11.4 Hz, 2H), 2.48 – 2.41 (m, 4H), 2.30 (s, 3H), 2.01 (q, *J* = 7.5 Hz, 2H), 1.93 – 1.85 (m, 2H), 1.58 (d, *J* = 12.6 Hz, 2H), 1.51 – 1.40 (m, 1H), 1.22 (qd, *J* = 12.3, 3.5 Hz, 2H), 1.02 (t, *J* = 7.4 Hz, 3H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 173.93, 158.75, 137.50, 135.42, 135.02, 129.33, 128.88, 128.71, 114.50, 55.65, 55.34, 53.86, 46.46, 42.66, 37.81, 32.09, 27.61, 20.89, 9.53. MS (ESI) m/z 379.2 (calcd 379.3 for $C_{25}H_{35}N_2O^+$ [M + H] $^+$).

5.1.12.4 N-(2-(4-(4-fluorobenzyl) piperidin-1-yl) ethyl)-N-(p-tolyl) propionamide (46)

Yield: 68%. Colorless oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.18 (d, J = 8.0 Hz, 2H), 7.09 – 7.03 (m, 4H), 6.97 – 6.91 (m, 2H), 3.85 – 3.76 (m, 2H), 2.87 (d, J = 10.4 Hz, 2H), 2.47 (t, J = 8.0 Hz, 4H), 2.37 (s, 3H), 2.02 (q, J = 7.4 Hz, 2H), 1.92 (t, J = 11.1 Hz, 2H), 1.57 (d, J = 12.5 Hz, 2H), 1.50 – 1.39(m, 1H), 1.24 (d, J = 14.4 Hz, 2H), 1.02 (t, J = 7.5 Hz, 3H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 173.81, 162.43, 160.01, 140.10, 137.65, 136.20, 130.36, 130.29, 130.11, 128.08, 114.93, 114.72, 55.64, 53.86, 46.41, 42.27, 37.84, 31.97, 27.71, 21.06, 9.60. MS (ESI) m/z 383.2 (calcd 383.2 for $C_{24}H_{32}FN_2O^+$ [M + H] $^+$).

5.1.12.5 N-(4-fluorophenyl)-N-(2-(4-(4-methylbenzyl) piperidin-1-yl) ethyl) propionamide (47)

Yield: 70%. Colorless oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.22 – 7.16 (m, 2H), 7.11 – 7.04 (m, 4H), 7.01 (d, J = 8.0 Hz, 2H), 3.79 (t, J = 7.1 Hz, 2H), 2.82 (d, J = 11.4 Hz, 2H), 2.48 – 2.40 (m, 4H), 2.31 (s, 3H), 2.05 – 1.96 (m, 2H), 1.88 (t, J = 10.8 Hz, 2H), 1.58 (d, J = 12.7 Hz, 2H), 1.46 – 1.38 (m, 1H), 1.26 – 1.14 (m, 2H), 1.03 (t, J = 7.4 Hz, 3H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 173.59, 162.92, 160.46, 138.79, 138.76, 137.49, 135.11, 130.19, 130.11, 128.93, 128.76, 116.42, 116.19, 55.70, 53.90, 46.47, 42.69, 37.84, 32.13, 27.75, 20.94, 9.51. MS (ESI) m/z 383.2 (calcd 383.2 for $C_{24}H_{32}FN_2O^+$ [M + H] $^+$).

5.1.12.6 N-(2-(4-(4-fluorobenzyl) piperidin-1-yl) ethyl)-N-(4-fluorophenyl) propionamide (48)

Yield: 65%. Yellow oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.22 – 7.13 (m, 2H), 7.09 – 7.04 (m, 4H), 6.98 – 6.93 (m, 2H), 3.79 (t, J = 7.1 Hz, 2H), 2.84 (d, J = 11.4 Hz, 2H), 2.50 – 2.39 (m, 4H), 2.05 – 1.96 (m, 2H), 1.89 (dd, J = 11.4, 10.0 Hz, 2H), 1.57 (d, J = 12.6 Hz, 2H), 1.51 –

1.38 (m, 1H), 1.27 – 1.14 (m, 2H), 1.03 (t, J = 7.4 Hz, 3H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 173.58, 162.91, 162.38, 160.44, 159.96, 138.75, 138.72, 136.14, 136.11, 130.32, 130.24, 130.17, 130.08, 116.41, 116.19, 114.88, 114.67, 55.66, 53.80, 46.42, 42.24, 37.84, 32.02, 27.74, 9.50. HRMS (ESI) m/z 387.2234 (calcd 387.2442 for $C_{23}H_{29}F_2N_2O^+$ $[M + H]^+$).

5.2 Receptor binding studies

5.2.1 Materials

The following specific radioligands and tissue sources were used: (a) σ1 receptor, [^{3}H]-(+)-pentazocine (250 μCi, Perkin-Elmer, NET-1056250UC), and guinea pig brain membranes; (b) μ-opioid receptor, [^{3}H]-diprenorphine (45.0 Ci/mmol; Perkin-Elmer), and human μ-opioid receptor-expressing CHO cells. Chemicals and reagents were purchased from different commercial sources and of analytical grade.

5.2.2 Membrane preparation

Guinea pig brain membrane preparation was performed by previously reported method [14]. Crude P2 membranes were prepared from frozen guinea pig brains minus cerebellum. Tissues were homogenized in ice-cold 10 mmol Tris-sucrose buffer (0.32 mol sucrose in 10 mmol Tris-HCl, pH 7.4) using an ULTRA TURAX homogenizer. The homogenates were centrifuged at 4°C at 1000 g for 10 min and the supernatant were saved. The resulting pellet was then resuspended in the same buffer, incubated for 10 min at 4°C, and centrifuged at 1000 g for 10 min. After that, the pellet was discarded and the supernatants were combined and centrifuging at 31 000 g for 15 min. The pellets were resuspended in 10 mL Tris-HCl, pH 7.4 in a volume of 3 mL/g and the suspension was allowed to incubate at 25°C for 30 min. Following centrifuging at 31 000 g for 15 min, the pellet was resuspended by gentle homogenization in 10 mmol Tris-HCl, pH 7.4 in a final volume of 1.53 mL/g tissue and aliquots were stored at -80°C until used.

Membranes from CHO-μ cells were prepared according to the published procedure [41]. CHO cells expressing the human μ-opioid receptor were stored at -80°C. The CHO cells were naturally thawed after being taken out in a -80°C refrigerator. Cells were homogenized in 50 mM Tris-HCl buffer (5 mM MgCl2 and 10% sucrose, pH 7.4) using an ULTRA TURAX homogenizer. The homogenates were centrifuged at 4°C at 1000g for 10 min, and the pellet was saved. The resulting pellet was then resuspended in the same buffer,

centrifuged at 50,000g, 4°C for 15 min. After that, the supernatants were discarded and the pellet was centrifuged three times in the same buffer at 50,000g, 4°C for 15 min. After centrifugation, the supernatants were discarded, and the pellet was stored at -80°C until use. The protein concentration of the suspension was determined by the method of Bradford [42]. Subsequently, the preparation contained 8 mg protein/mL.

5.2.3 General Procedures for the Binding Assays.

All the test compounds were prepared by dissolving in 5% DMSO. The filter mats were presoaked in 0.5% polyethylenimine solution for 2 h at room temperature before use. The following specific radioligands and tissue sources were used: (a) sigma-1 receptor, [^{3}H]-(+)-pentazocine, guinea pig brain membranes; (b) μ-opioid receptor, [^{3}H]-diprenorphine, human μ-opioid receptor- expressing CHO cells.

The total binding (TB) was determined in the absence of nonspecific binding and compounds while specific binding (SB) was determined in the presence of compounds. Nonspecific binding (NB) was determined as the difference between total and specific binding. Nonspecific binding was determined in the presence of 10μmol haloperidol. Percentage of inhibition (%) was calculated as the following equation: Percentage of inhibition (%)=(TB-SB)/(TB-NB) x 100%. Blank experiments were carried out to determine the effect of 5% DMSO on the binding and no effects were observed. Compounds were tested at least three times over a six-concentration range (10^{-5} mol to 10^{-10} mol), IC_{50} values were determined by nonlinear regression analysis using Hill equation curve fitting. K_i values were calculated based on the Cheng and Prussoff equation: $K_i = IC_{50} / (1+C / K_d)$. In the equation, C represents the concentration of the hot ligand used and K_d its receptor dissociation constant were calculated for each labeled ligand. Mean K_i values and SEM are reported for at least three independent experiments [43].

5.2.4 Sigma-1 receptor binding assays

Binding of [^{3}H]-(+)-pentazocine at σ1 receptor was performed according to D. L. Dehaven-Hudkins et al [44].with minor modifications[14]. The binding properties of the test compounds to guinea pig σ1 receptor were studied in guinea pig brain membranes using [^{3}H]-(+)-pentazocine as the radioligand. For each total binding assay tube were added 900 μL of the tissue suspension, 50 μL of 4.0 nM [^{3}H]-(+)-pentazocine, 50 μL Tris-HCl

buffer, pH 8.0. For nonspecific binding, to each assay tube was added 900 µL of the tissue suspension, 50 µL of [^{3}H]-(+)-pentazocine, and 50 µL of 10 µM haloperidol. For specific binding, to each assay tube was added 900 µL of the suspension, 50 µL of [^{3}H]-(+)-pentazocine, 50 µL of reference drug or test compounds solution in various concentrations (10^{-5} mol to 10^{-10} mol). The tubes were incubated at 25°C for 150 min. The incubation was followed by rapid vacuum filtration through Whatman GF/C glass filters, and the filtrates were washed twice with 5 mL of cold buffer and transferred to scintillation vials. Scintillation fluid (2.0 mL) was added, and the radioactivity bound was measured using a Beckman LS 6500 liquid scintillation counter.

5.2.5 Mu-opioid receptor binding assays

Binding of [^{3}H]-diprenorphine at µ-opioid receptor was performed according to M. Spetea et al [41]. with minor modifications [14]. A suspension of membranes from human µ-opioid receptor-expressing CHO cells in 50 mM Tris-HCl buffer (pH 7.4) containing 5 mM $MgCl_2$ and 10% sucrose was used. For total binding, to each assay tube was added 900 µL of the tissue suspension, 50 µL of 4.0 nM [^{3}H]-diprenorphine, and 50 µL of Tris-HCl buffer. For nonspecific binding, to each assay tube was added 900 µL of the tissue suspension, 50 µL of [^{3}H]-diprenorphine, and 50 µL of 10 µM DAMGO. For specific binding, to each assay tube was added 900 µL of the suspension, 50 µL of [^{3}H]-diprenorphine, 50 µL of reference drug or test compounds solution in various concentrations (10^{-5} mol to 10^{-10} mol). The tubes were incubated at 25°C for 150 min. The incubation was followed by rapid vacuum filtration through Whatman GF/C glass filters, and the filtrates were washed twice with 5 mL of cold buffer and transferred to scintillation vials. Scintillation fluid (2.0 mL) was added, and the radioactivity bound was measured using a Beckman LS 6500 liquid scintillation counter.

5.3 In vivo test

5.3.1 Animals

Sprague-Dawley (SD) Rats. (230-280 g) and ICR mice (23-35g) were used as experimental animals in this study. All the animals were housed under standardized conditions for light, temperature and humidity, received standard rat chow and tap water and libitum. Animals were assigned to different experimental groups randomly, each kept in

a separate cage. All research-involving animals in this study follow the guidelines of the byelaw of experiments on animals. In addition, have been approved by the Ethics and Experimental Animal Committee of Jiangsu Nhwa Pharmaceutical Co., Ltd.

5.3.2 Acute toxicity

Mice (10 mice for each group) were subcutaneous administration of a 4 mL/kg volume of vehicle 0.5% methylcellulose (Sigma-Aldrich) or increasing dose of test compounds (50, 76, 115, 174, 264, 400 mg/kg). The number of surviving animals was recorded after 24h of drug administration. The percent mortality in each group was calculated. The LD_{50} values were calculated by using Statistical Package for Social Sciences (SPSS) program.

5.3.3 Formalin test

Formalin tests were performed as described by Cendán et al [45]. With minor modifications [14, 32]. A diluted formalin solution (100 μL of a 5% formalin solution, 1.85% formaldehyde) was intraplantar injection into the dorsal surface of the right hind paw of the rats [46]. Immediately thereafter, the mouse was put into a glass cylinder and the observation period started. A mirror was placed behind the glass cylinder to allow clear observation of the paws. Nociceptive behavior induced by formalin was quantified as the time spent licking or biting the injected paw during two different periods individually recorded. The first period was recorded 0-5 min after the injection of formalin and was considered indicative of formalin-evoked nociception phase I; the second period was recorded 15-45 min after formalin injection and was considered indicative of formalin evoked nociception phase II. The mice (n=10 per group) received i.p. administration of a 4 mL/kg volume of vehicle 30% PEG 400 (Sigma-Aldrich) or test compound (s.c.) 30 min before intraplantar (ipl) formalin injection.

The anti-nociceptive effect induced by the different treatments in the formalin test was calculated by the following formula: antinociceptive effect (%) = $(LT_V\text{-}LT_D)/LT_V \times 100\%$, where LT_v represent the licking time in vehicle injected animals, LT_D means licking time in drug-injected animals.

5.3.4 CCI Model

The chronic constriction injury of the sciatic nerve (CCI) as an animal model was used to study the efficacy of treatments in neuropathic pain. Rats were randomly separated into

several groups: sham control and vehicle- and drug-treated groups, and each group contained 10 rats. Pain threshold basal values of each group were measured 2 days before surgery. The pain thresholds were measured again 14 days after surgery to check the model was successful or not. Drugs single dose were subcutaneous injection administration of a 0.2ml/100g volume and vehicle 30% PEG400 (Sigma-Aldrich). Each group, the paw withdrawal threshold was measured 30 minutes after a single administration, and the maximum analgesic effect MPE% after administration was calculated by the following formula: MPE%=[(test latency-base line latency)/(50- base line latency)]×100% [47].

The CCI of the sciatic nerve surgery was performed as described by Bennett and Xie [33]. With minor modifications [14]. The SD rats were anesthetized with 7% chloral hydrate (350mg/kg) and the right common sciatic nerve was exposed at the level of the mid-thigh of the right hind paw. At about 1 cm proximal to the nerve trifurcation, about 7 mm of the nerve was freed and four ligatures (4.0 silk tread) were tight loosely with a distance of ca. 1.0 mm. The nerve was only barely constricted. The muscle was than stitched and the skin incision closed with wound clips. The rats with sciatic nerve exposure without ligation served as the sham control group.

The von Frey test was performed as described by Chaplan [48]. With minor modifications. Briefly, animals were placed in a transparent test chamber with a wire-mesh grid floor through which von Frey monofilaments were applied. The monofilaments were applied in increasing force until the rats withdrew the ipsilateral, nerve injury paw using an up−down paradigm. Clear paw withdrawal, sharking, or licking was considered as nociceptive-lick response to determine the mechanical withdrawal threshold (MWT). Animals were adapted to the testing situation for at least 30 min before the sessions started. For each measurement, at least 10 min elapsed between, the paw was sampled three times, and a mean calculated.

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Supporting material

Supporting information available: ^{1}H NMR, ^{13}C NMR, HRMS, and HPLC of compounds **44** and **48**; additional receptors binding affinities of compound **44** and **48**.

Abbreviations used

σ_1, sigma-1; σ_2, sigma-2; SAR, structure activity relationship; CCI, chronic constriction injury; NMDA, N-methyl-D-aspartate; 5-HT_{1A}, serotonin-1A; 5-HT_{2A}, serotonin-2A; H_3, histamine-3; α_1, adrenergic-1; α_2, adrenergic-2; CB_1, Cannabinoid-1; CB_2, Cannabinoid-2; ED_{50}, 50% effective dose; LD_{50}, median lethal dose.

Notes

The authors declare no competing financial interest.

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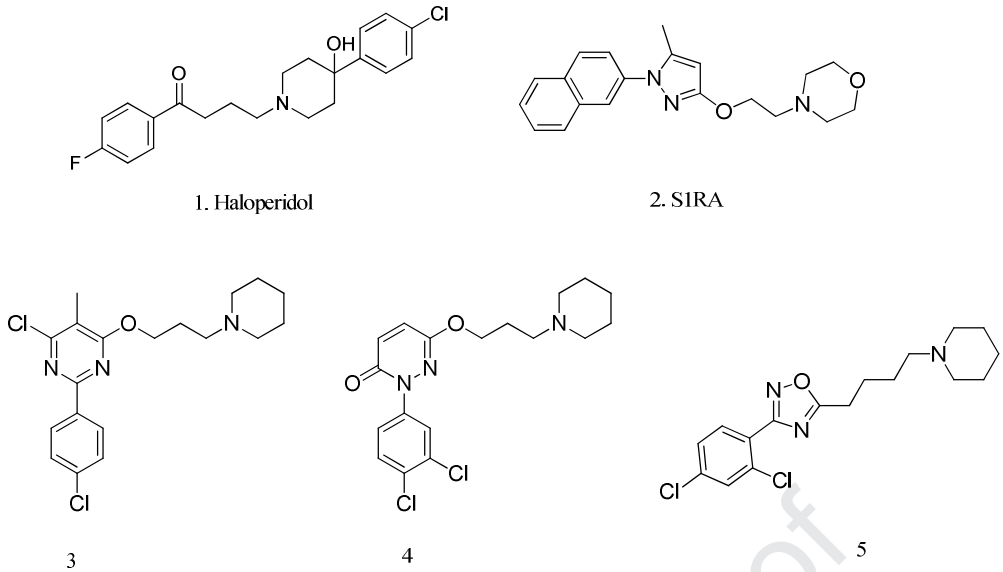
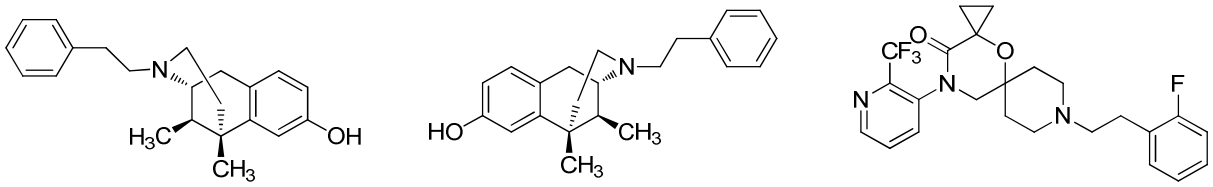


Fig.1. Representative sigma-1 receptor antagonists.



6a. (-)-Phenazocine
K_i μ = 55 nM
K_i σ_1 = 3.90 nM

6b. (+)-Phenazocine
K_i μ = 0.2 nM
K_i σ_1 = 61.7 nM

7
K_i μ = 175 nM
K_i σ_1 = 58 nM

Fig.2. Chemical structures of ligands with μ receptor agonist/σ_1 receptor antagonists profile

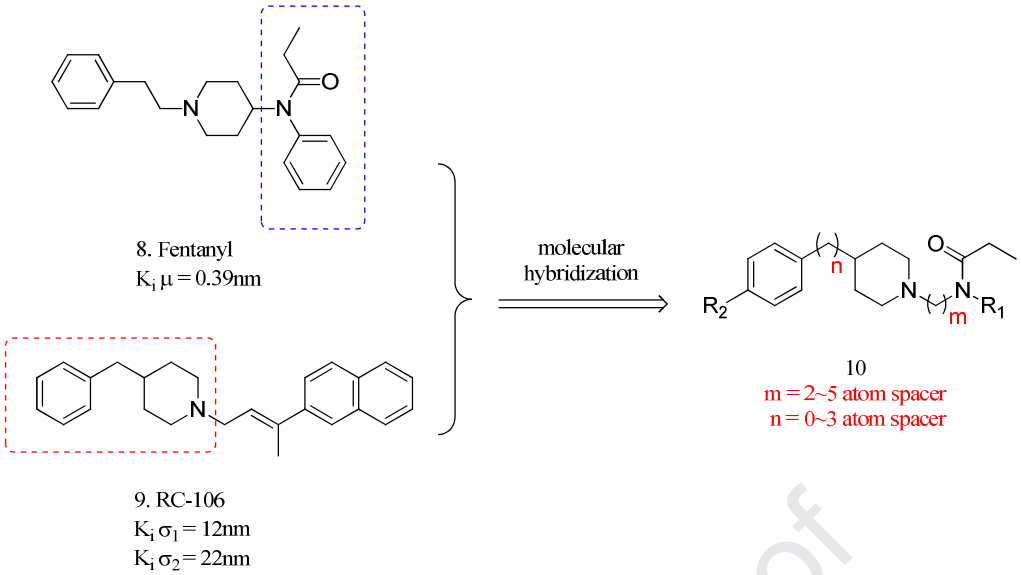
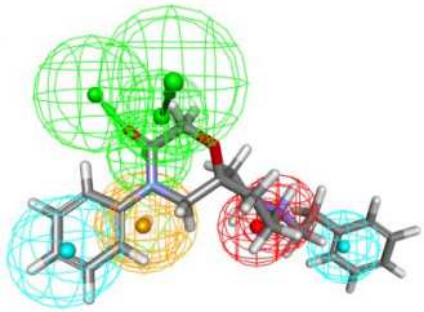
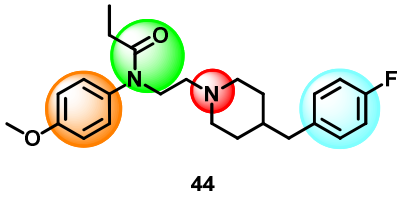


Fig.3. Design of new piperidine propionamide derivatives

A B





44

Fig.4. Mónica García pharmacophoric σ_1 receptor antagonist/μ receptor agonist model (A) and chemical structure of compound 44(B).

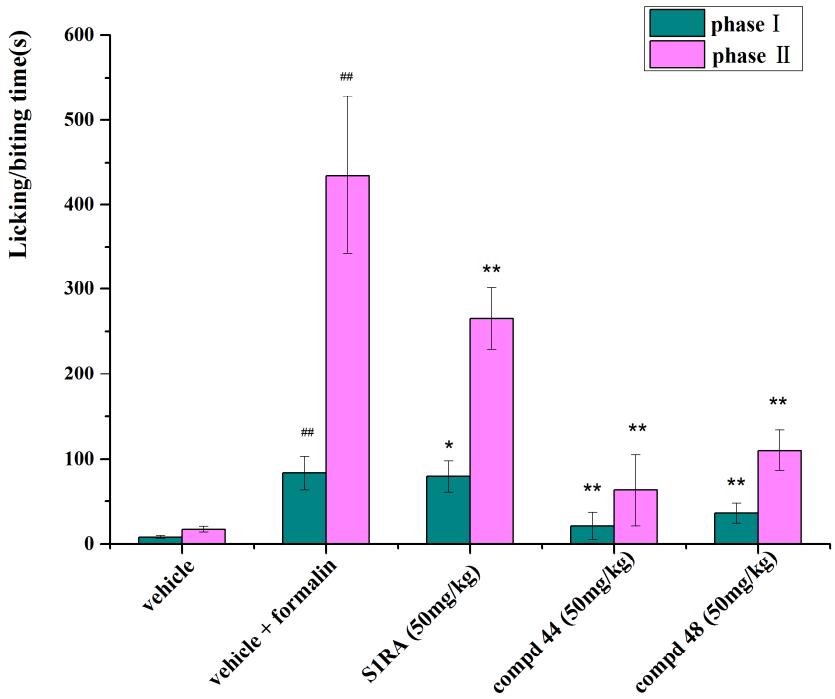


Fig.5. Anti-nociceptive effect of S1RA (50 mg/kg), compound 44 (50 mg/kg) and 48 (50 mg/kg) in phase I (0-5 min) and phase II (15-45 min) of the formalin test. Each column and vertical line represents mean ± SEM of the values obtained in at least 10 animals. Statistically significant differences: #p < 0.05; ##p < 0.01 vs. vehicle; *p < 0.05; **p < 0.01 vs. vehicle + formalin (two-way ANOVA followed by Newman-Keuls test).

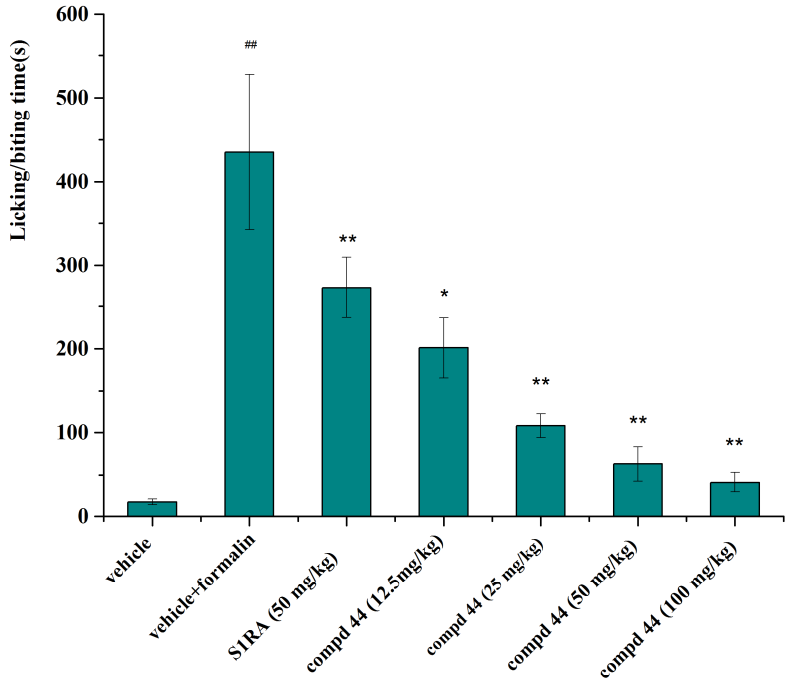


Fig.6. Anti-nociceptive effect of S1RA (50 mg/kg), compound 44 in phase II (15-45 min) of the formalin test. Each column and vertical line represents mean±SEM of the values obtained in at least 10 animals. Statistically significant differences: #p < 0.05; ##p < 0.01 vs vehicle; *p < 0.05; **p < 0.01 vs vehicle + formalin (two-way ANOVA followed by Newman-Keuls test).

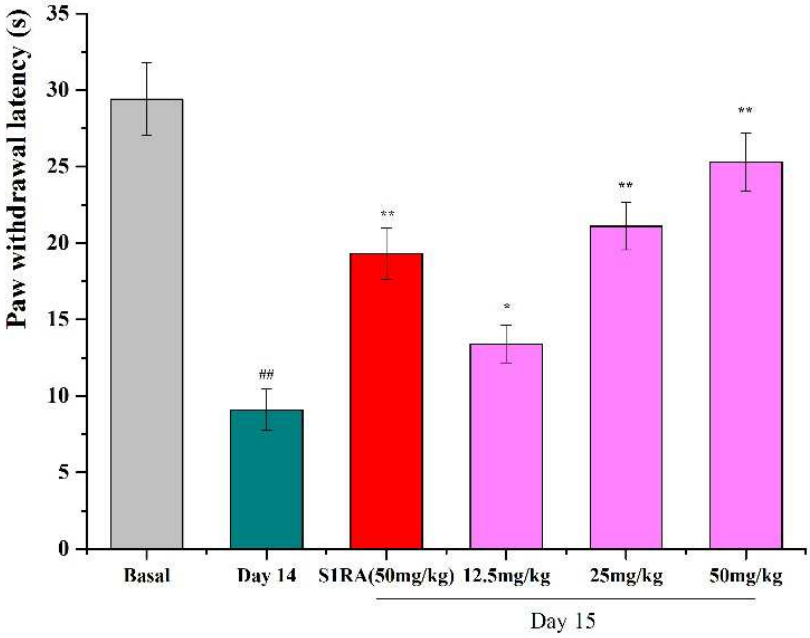
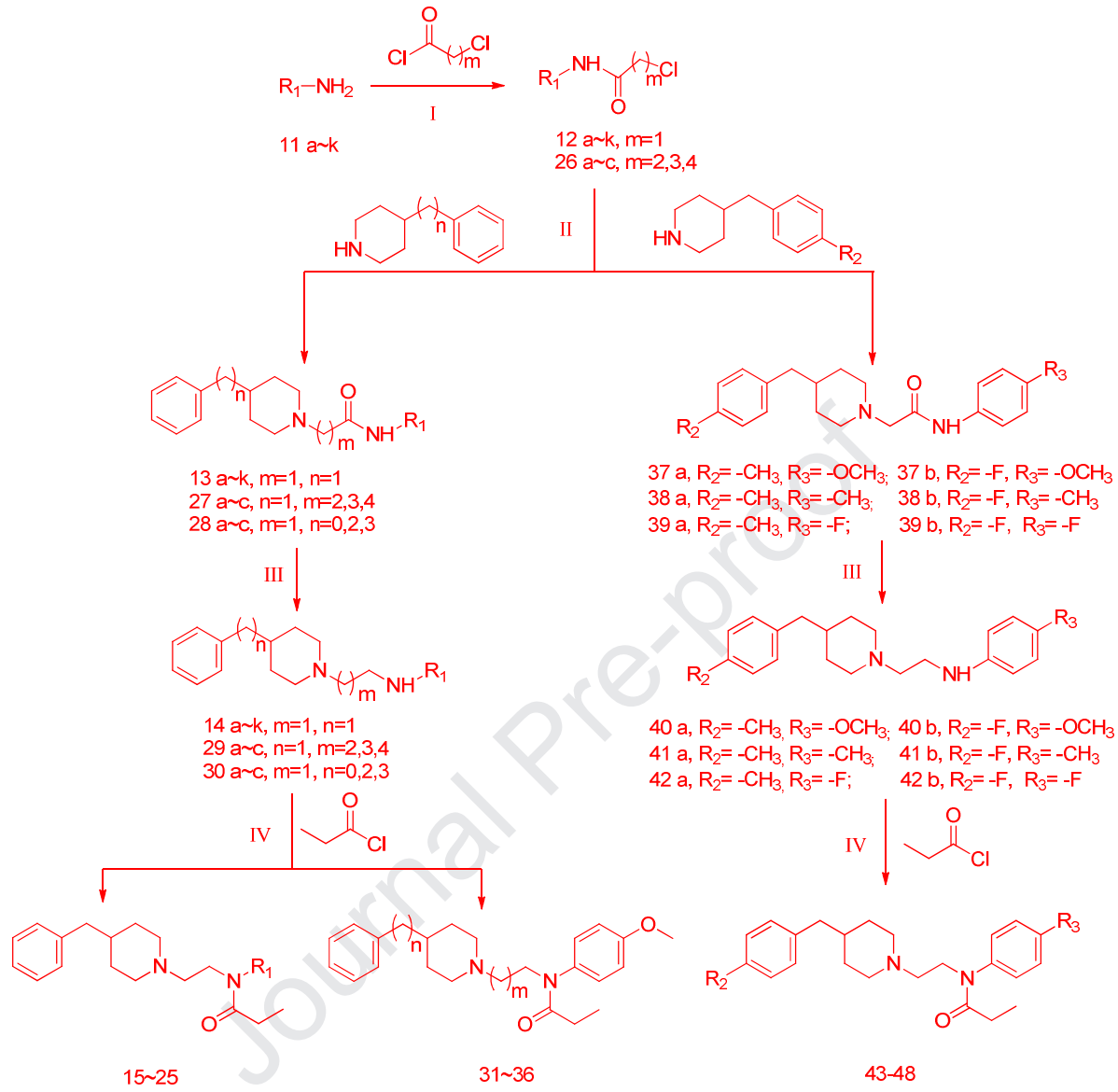


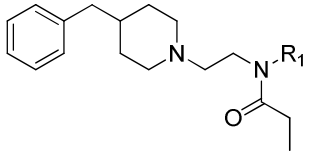
Figure 7. Antiallodynic effects of compound 44 on the expression of neuropathic pain in CCI model (Mechanical allodynia). Date obtained from 10 rats per group and expressed as mean ± SEM pressure threshold (g) evoking paw withdrawal or latency (s) to paw withdrawal. Statistically significant differences between the rats before surgery (basal) and surgery groups: * $p < 0.05$, ** $p < 0.01$ vs vehicle; # $p < 0.05$; ## $p < 0.01$ vs basal treatment on day 14 (Two-Way ANOVA followed by Newman−Keuls test).

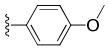
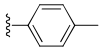
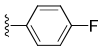
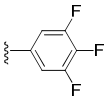
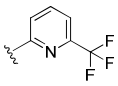
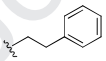
Scheme 1



Reagents and conditions: (I) K_2CO_3, acetone, rt, 4h; (II) K_2CO_3, acetone, rt, 8h; (III) $LiAlH_4$, THF, 65°C, 4h; (IV) Et_3N, DCM, 0°C, 3h.

Table 1 Binding affinities for the σ_1 and μ receptors of compounds 15~25.



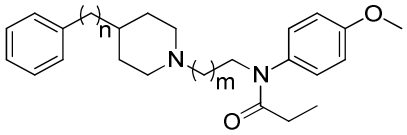
Compound	R_1	K_i σ_1(nM)[a]	K_i μ(nM)[b]
15		38.6 ± 1.7[c]	3.7 ± 0.23
16		1.5 ± 0.07	20.4 ± 1.1
17		1.6 ± 0.22	16.7 ± 0.94
18		4.1 ± 0.35	19.4 ± 0.72
19		573 ± 37	>2000
20		402 ± 34	238 ± 28
21		506 ± 28	253 ± 12
22		835 ± 132	>2000
23		17.4 ± 1.9	694 ± 49
24		18.7 ± 0.9	573 ± 89
25		269 ± 18	>2000

[a] Affinities were determined in guinea pig brain using [^{3}H]-(+)-pentazocine.

[b] Affinities were determined in CHO cell using [^{3}H]-DAMGO.

[c] K_i values are given as mean ± SD of three independent experiments.

Table 2 Binding affinities for the σ_1 and μ receptors of compounds 16 derivatives.



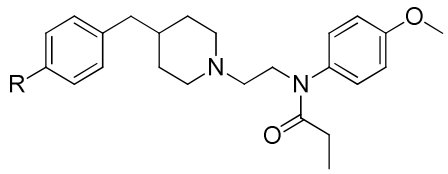
compound	n	m	K_i σ_1(nM)[a]	K_i μ(nM)[b]
16	1	1	1.5 ± 0.07[c]	20.4 ± 1.1
31	1	2	2.2 ± 0.16	43 ± 3.4
32	1	3	18 ± 1.92	78 ± 6.3
33	1	4	42 ± 4.8	165 ± 2.4
34	0	1	39 ± 4.7	26 ± 1.7
35	2	1	372 ± 32	82 ± 9.3
36	3	1	893 ± 74	74 ± 3.1

[a] Affinities were determined in guinea pig brain using [^{3}H]-(+)-pentazocine.

[b] Affinities were determined in CHO cell using [^{3}H]-DAMGO.

[c] K_i values are given as mean ± SD of three independent experiments.

Table 3 Binding affinities for the σ_1 and μ receptors of compounds 43 and 44.



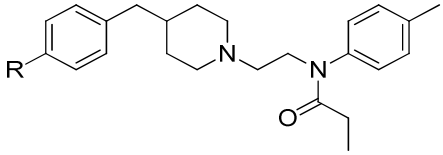
Compound	R	K_i σ_1(nM)[a]	K_i μ(nM)[b]
16	H	1.5 ± 0.07[c]	20.4 ± 1.1
43	CH_3	35 ± 1.6	77 ± 7.4
44	F	1.86 ± 0.05	2.1 ± 0.16

[a] Affinities were determined in guinea pig brain using [^{3}H]-(+)-pentazocine.

[b] Affinities were determined in CHO cell using [^{3}H]-DAMGO.

[c] K_i values are given as mean ± SD of three independent experiments.

Table 4 Binding affinities for the σ_1 and μ receptors of compounds 45 and 46.



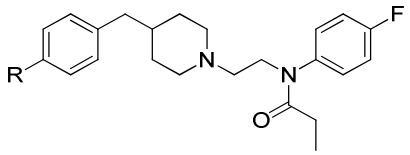
Compound	R	K_i σ_1(nM)[a]	K_i μ(nM)[b]
17	H	1.6 ± 0.22	16.7 ± 0.94
45	CH_3	29 ± 1.3	82 ± 19
46	F	2.4 ± 0.32	27.9 ± 3.8

[a] Affinities were determined in guinea pig brain using [^{3}H]-(+)-pentazocine.

[b] Affinities were determined in CHO cell using [^{3}H]-DAMGO.

[c] K_i values are given as mean ± SD of three independent experiments.

Table 5 Binding affinities for the σ_1 and μ receptors of compounds 47 and 48.



Compound	R	K_i σ_1(nM)[a]	K_i μ(nM)[b]
18	H	4.1 ± 0.35	19.4 ± 0.72
47	CH_3	47 ± 2.4	148 ± 26
48	F	1.3 ± 0.22	5.6 ± 0.36

[a] Affinities were determined in guinea pig brain using [^{3}H]-(+)-pentazocine.

[b] Affinities were determined in CHO cell using [^{3}H]-DAMGO.

[c] K_i values are given as mean ± SD of three independent experiments.

Table 6 Binding affinities for the additional receptors of compounds 44 and 48.

Receptor/ ion channel	Compound 44	Compound 48
	Inhibition (%)[a]	
σ_2	29.13 ± 1.2	31.75 ± 0.2
H_3	34.64 ± 3.1	9.75 ± 0.5
5-HT_{1A}	45.18 ± 0.6	23.04 ± 2.5
5-HT_{2A}	43.62 ± 2.1	34.42 ± 0.8
5-HT transporter	10.92 ± 0.3	44.60 ± 1.7
Noradrenaline transporter	18.53 ± 0.4	28.81 ± 1.3
CB_1	20.83 ± 1.6	17.36 ± 2.7
CB_2	32.64 ± 2.8	19.89 ± 0.6
α_1	24.74 ± 2.4	39.92 ± 4.3
α_2	45.87 ± 1.4	32.31 ± 2.9
NMDA	14.32 ± 0.4	20.46 ± 0.7

[a] % inhibition was determined at the concentration of 1 μM.

1 **Table 7 LD_{50} value of compounds 44 and 48.**

compound	LD_{50}(mg/kg)[a]
S1RA	357.4 (312.6−418. 5)
44	396.7 (347.8−433. 2)
48	415.8 (375.3−538. 6)

2 [a] 95% confidence limits.

Highlights

- 1. A new series of piperidine propionamide derivatives was designed and synthesized.
- 2. Compound **44** showed highest affinities to σ_1 receptor and μ receptor with mixed σ_1/μ receptor profiles.
- 3. Compound **44** performed a dose-dependent analgesic effect in the formalin test.
- 4. Compound **44** performed equivalent analgesic effect with S1RA.

Declaration of interests

☑ The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

☐ The authors declare the following financial interests/personal relationships which may be considered as potential competing interests: